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Transcriptome Analysis Applied to Survival of *Shewanella oneidensis* MR-1 Exposed to Ionizing Radiation

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The ionizing radiation (IR) dose that yields 20% survival (D_{20}) of *Shewanella oneidensis* MR-1 is lower by factors of 20 and 200 than those for *Escherichia coli* and *Deinococcus radiodurans*, respectively. Transcriptome analysis was used to identify the genes of MR-1 responding to 40 Gy (D_{20}). We observed the induction of 170 genes and repression of 87 genes in MR-1 during a 1-h recovery period after irradiation. The genomic response of MR-1 to IR is very similar to its response to UV radiation (254 nm), which included induction of systems involved in DNA repair and prophage synthesis and the absence of differential regulation of tricarboxylic acid cycle activity, which occurs in IR-irradiated *D. radiodurans*. Furthermore, strong induction of genes encoding antioxidant enzymes in MR-1 was observed. DNA damage may not be the principal cause of high sensitivity to IR, considering that MR-1 carries genes encoding a complex set of DNA repair systems and 40 Gy IR induces less than one double-strand break in its genome. Instead, a combination of oxidative stress, protein damage, and prophage-mediated cell lysis during irradiation and recovery might underlie this organism's great sensitivity to IR.

Ionizing radiation (IR) is potentially lethal and mutagenic to all organisms. The cellular response to IR is complex due to the variety of targets in a cell. IR can damage cellular components through direct deposition of radiation energy into biomolecules and also indirectly by generating reactive oxygen species (ROS). Hydrogen peroxide (H_2O_2) and hydroxyl radicals (HO·) are major oxidizing species produced by the radiolysis of water, and superoxide ions ($O_2^{\cdot -}$) are formed in the presence of dissolved oxygen (8, 24, 25, 27, 28).

Generally, the cytotoxic and mutagenic effects induced by IR are thought to be the result of DNA damage caused during the course of irradiation, which includes single-strand breaks (SSB), double-strand breaks (DSB), base modification, abasic sites, and sugar modification (11, 27). The amounts of DNA damage caused by given doses of IR for different bacteria are very similar, although the range of IR resistance levels is large. The dissimilatory metal ion-reducing *Shewanella oneidensis* strain MR-1 is extremely sensitive to IR: e.g., 90% of MR-1 cells are killed by less than one DSB in the genome (4). *S. oneidensis* is also one of the most UV- and desiccation-sensitive organisms reported (4, 22). The IR resistance of MR-1 is about 20 times less than that of *Escherichia coli* and about 200 times less than that of *Deinococcus radiodurans* (4). It is unclear why *S. oneidensis* MR-1 is exceptionally sensitive to radiation.

Since more than 80% of IR energy deposited in cells results in the ejection of electrons from water, a large amount of ROS is produced in cells exposed to IR (8, 25). Although both H_2O_2 and $O_2^{\cdot -}$ are relatively stable, HO· are extremely toxic to cells due to their very high reactivity (25). In cells, it appears that proteins are the first major class of molecules damaged by IR-induced ROS (6, 21). ROS can also be generated during cell metabolism when electrons leak from the substrate side of the respiratory chain (14, 26). The autoxidation of flavoenzymes yields a mixture of $O_2^{\cdot -}$ and H_2O_2 (20). Several [4Fe-4S] cluster-containing proteins have been reported to be exceptionally sensitive to these ROS. For example, the dehydratase aconitase (AcnB) contains a [4Fe-4S] cluster that is readily oxidized by $O_2^{\cdot -}$ in vivo, resulting in enzyme deactivation and the release of bound ferrous iron [Fe(II)] (14). Subsequent oxidation of Fe(II) by H_2O_2 (Fenton reaction) generates HO·, which attacks all biomolecules (6, 16, 27). Recently, an association between intracellular Mn/Fe concentration ratios and bacterial IR resistance was reported, where very high and very low Mn/Fe ratios correlated with very high and very low levels of resistance, respectively (4). The high Mn content of radiation-resistant bacteria might protect cells from ROS (4, 10). For example, intracellular Mn engaged in Mn(II,III) redox cycling is potentially a powerful scavenger of H_2O_2 and $O_2^{\cdot -}$ (1). In contrast, high intracellular Fe may cause a proliferation of ROS during recovery and promote the production of highly toxic HO· by Fenton-type chemistry (14, 28). Thus, Fe-rich, Mn-poor bacteria such as *Shewanella* spp. might be predisposed to oxidative stress produced both during and after irradiation (4, 10, 14).

Comparative genomic analyses support the view that *S. oneidensis* encodes a complex set of DNA repair and protection functions, which have functional homologs in radiation-resistant prokaryotic species (4, 10). A systematic genome-wide examination of the genes and pathways involved in recovery would be useful for a further understanding of how radiation

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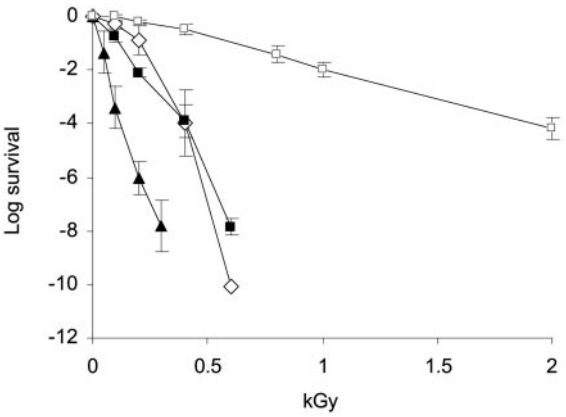

FIG. 1. Survival of *Shewanella* species exposed to acute gamma radiation (^{60}Co). Solid triangles, *S. oneidensis* MR-1 (ATCC 700550); solid squares, *S. putrefaciens* CN-32 (ATCC BAA-453); open diamonds, *S. amazonensis* (ATCC 700329); open squares, *E. coli* K-12 (MG1655). Strains were grown to an OD_{600} of 0.9 and irradiated on ice as previously described (4).

affects *S. oneidensis* MR-1. Here we report the analysis of genomic expression within *S. oneidensis* MR-1 cells recovering from 40 Gy IR using a whole-genome DNA microarray. We find that the hallmark components of *S. oneidensis* recovery from IR encompass responses observed in other organisms, including induction of the error-prone DNA repair (SOS) response, genes for homologous recombination, and uncharacterized genes. Unlike the response of *D. radiodurans* to IR (18), irradiated *S. oneidensis* did not suppress tricarboxylic acid cycle (TCA) activity and strongly induced genes involved in fighting against ROS and synthesizing prophages.

Survival of *Shewanella* species after exposure to IR. The IR resistance profile of *S. oneidensis* MR-1 (ATCC 700550) was previously reported (4). To determine whether or not other *Shewanella* species are also IR sensitive, *Shewanella amazonensis* (ATCC 700329) and *Shewanella putrefaciens* CN-32 (ATCC BAA-453) were tested for their resistance to acute IR (Fig. 1). *E. coli* K-12 was used as a reference. When grown in TGY (tryptone, glucose, yeast extract) medium, the IR doses yielding 10% (D_{10}) CFU survival for the strains were as follows: *S. oneidensis*, ~70 Gy; *S. putrefaciens*, ~100 Gy; *S. amazonensis*, ~200 Gy; and *E. coli* K-12, ~700 Gy (Fig. 1). Thus, strain MR-1 is representative of the extreme IR sensitivity of *Shewanella* spp. Our previous transcriptional profiling studies on MR-1 recovering from UV (254 nm) radiation used cells grown in Davis medium (Difco) to an optical density at 600 nm (OD_{600}) of 0.2, with 15 mM lactate as the sole carbon source (23). Therefore, we determined the IR resistance profile of MR-1 when it was grown in Davis medium to an OD_{600} of 0.2. MR-1 showed similar sensitivity to IR when grown in TGY medium; the D_{20} (20% survival) value of MR-1 in Davis medium was 40 Gy.

Microarray experiment, quality of microarray hybridization data, and data analysis. The *S. oneidensis* MR-1 whole-genome cDNA array containing about 95% of total *S. oneidensis* MR-1 open reading frames (9) was used to examine the transcriptome expression dynamics of MR-1 over a period of 1 h following IR exposure. Briefly, 80 ml of MR-1 culture grown in Davis medium to an OD_{600} of 0.2 was divided in half. Half of the culture (40 ml) was irradiated on ice to a total dose of 40 Gy (^{60}Co, 109 Gy/min; Gammacell irradiation unit, model 109; J. L. Shepard and Associates). The nonirradiated control culture was incubated on ice for the same length of time (about 20 s) as the culture being irradiated. After irradiation, the irradiated cell culture (40 ml) and nonirradiated control culture (40 ml) were each transferred to a separate 100-ml flask (without dilution into fresh medium) and incubated at 30°C on a shaker. An aliquot of cells (12 ml) from each flask was collected after 5, 20, and 60 min of incubation for RNA extraction as described previously (22, 23). Three independent cell cultures and irradiation treatments were performed and served as biological replicates for gene expression experiments. RNA extraction, probe labeling, microarray hybridization, image acquisition, and processing were carried out as described previously (22, 23). For each time point, two technical replicates in hybridization (fluorescent dye reversal) were carried out for each biological replicate. Thus, six data points were available for each time point and enabled the use of statistical tests to determine significant changes in gene expression.

The gene expression ratios of the irradiated samples to the respective nonirradiated control were normalized using the pooled-common-error model provided by the statistical analysis software ArrayStat v. 2.0 (Imaging Research Inc., Ontario, Canada). The outliers, represented by the data points that were not consistently reproducible and had a disproportionately large effect on the statistical result, were removed. The standard *t* test was performed so that a two-tailed probability of a mean deviating from 1.0 could be calculated and used to determine the significance for each data point. Genes that showed a statistically significant difference in expression ($P < 0.05$) and were >2-fold in magnitude as up-regulated and <1.5-fold as down-regulated were analyzed further. Identification of groups of genes exhibiting similar expression patterns was performed using the pairwise average-linkage hierarchical clustering algorithm provided by the Cluster software (http://rana.stanford.edu/). The complete microarray data set for the recovery time course is listed in supplemental Tables 2 and 3 (posted on our website at http://cme.msu.edu/tiedjelab/gamma). The raw data set is also available at our website.

Quantitative reverse transcription-PCR for seven selected genes was used to test the robustness of the microarray hybridization data, which included two highly induced genes (*recA* and *katB*), a moderately induced gene with high variations (SO1762), a slightly induced gene (*trxC*), a slightly repressed gene (SO1490), and two genes with no changes in expression level (SO1924 and SOA0154), as described previously (23). *ldhA* was used as an internal control to normalize the difference in reverse transcription efficiency. A high correlation ($R^2 = 0.9115$) was obtained between microarray analysis and quantitative reverse transcription-PCR analysis (supplemental Table 1 on our website).

General patterns of expression in response to ionizing radiation. A total of 170 genes were induced and 87 genes were repressed at least at one time point examined during the 1-h recovery period. Similar to the gene expression profile of MR-1 following UVC irradiation (23), the highest differential expression in response to IR appeared at 60 min, with 147 genes induced and 46 genes repressed (Fig. 2). Of the total of

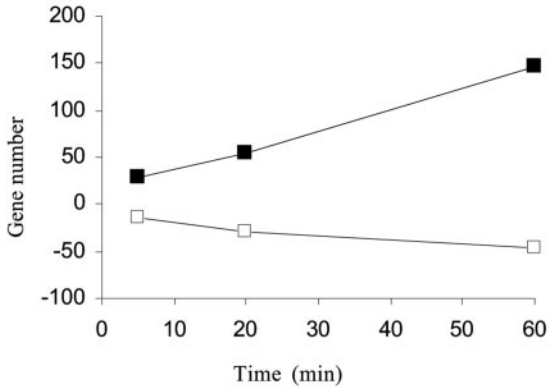

FIG. 2. Global gene expression trend in response to IR during a 1-h recovery period in *S. oneidensis* MR-1. Solid squares represent up-regulated genes, and open squares represent down-regulated genes. A positive number on the *y* axis represents the number of up-regulated genes, and a negative number represents the number of down-regulated genes.

170 IR-induced genes, 100 were phage-related genes, which was the largest group among IR-induced genes (Fig. 3, phage). Sixty up-regulated genes were distributed into 14 COG functional categories, of which the top five categories were L (DNA replication, recombination, and repair; 11 genes), R (general function prediction only; 9 genes), O (posttranslational modification, protein turnover, and chaperones; 7 genes), P (inorganic ion transport and metabolism; 6 genes), and S (function unknown; 6 genes) (Fig. 3). A great number of genes involved in DNA repair (K and L) and defending against oxidative stress (O and P) were up-regulated in MR-1 following IR irradiation, which indicates IR induces both DNA damage and oxidative stress in MR-1 at 40 Gy.

Of the 87 down-regulated genes, 17 genes coded for hypothetical proteins (Fig. 3, not in COG) and 70 genes were distributed into 16 COG functional categories (Fig. 3). The largest group was P, containing 16 genes that are involved in inorganic ion transport and metabolism. Interestingly, all genes involved in sulfate reduction were down-regulated within 20 min postirradiation (supplemental Table 3 on our website). In addition, genes involved in molybdenum transport (*modA*, *modB*, *modC*, and SO3967) were all significantly repressed at 5 min following irradiation (supplemental Table 3 on our website). The second largest group was E, containing eight genes involved in amino acid transport and metabolism (supplemental Table 3 on our website). Repression of both inorganic ion and amino acid transport and metabolism can decrease the energy requirement in IR-irradiated cells and thus minimize the generation of additional ROS. However, in contrast to what was observed for irradiated *D. radiodurans* (18), we did not observe any repression of genes participating in the TCA cycle nor the induction of genes involved in glyoxylate bypass.

TABLE 1. Induction of prophage-related genes following IR and UVC irradiation

Prophage	Total no. of genes[a]	Total no. of genes	
		IR induced	UVC induced
LambdaSo	75	65	56
MuSo1	42	18	15
MuSo2	53	17	16

[a] According to Heidelberg et al (12).

Induction of prophage-related genes. There are three prophages in the MR-1 genome (12). A 51,857-bp lambda-like phage genome (75 lambdaSO genes) is integrated in the *S. oneidensis* MR-1 genome and is also present in nonintegrated form. Additionally, two phylogenetically distinct phages (MuSo1 and MuSo2) related to *Escherichia coli* Mu have been identified, carrying 42 genes and 53 genes, respectively (12). Totals of 65, 18, and 17 open reading frames of prophage LambdaSo, MuSo1, and MuSo2 were up-regulated in a synchronous, function-specific manner, respectively (Table 1; and see supplemental Table 2, on our website), which indicates that, similar to UV, IR activates

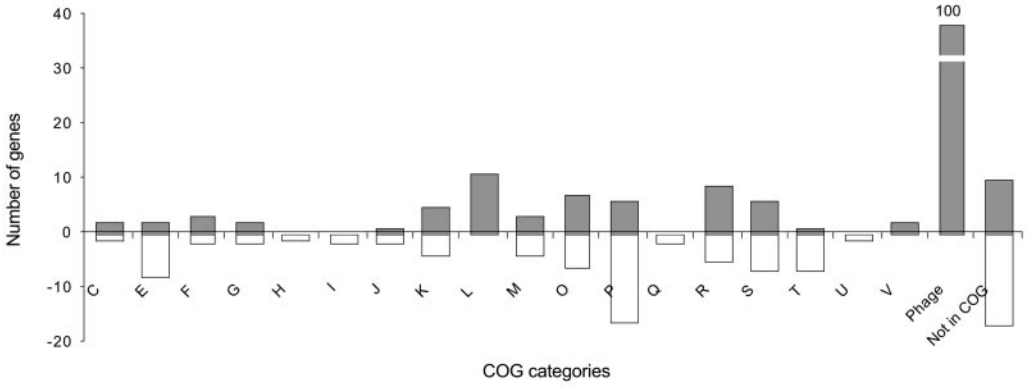

FIG. 3. Functional distribution of the differentially expressed genes in COG's functional categories. The total number of up-regulated genes is 170, and the total number of down-regulated genes is 87. C, energy production and conversion; E, amino acid transport and metabolism; F, nucleotide transport and metabolism; G, carbohydrate transport and metabolism; H, coenzyme metabolism; I, lipid metabolism; J, translation, ribosomal structure, and biogenesis; K, transcription; L, DNA replication, recombination, and repair; M, cell envelope biogenesis, outer membrane; N, cell motility and secretion; O, posttranslational modification, protein turnover, and chaperones; P, inorganic ion transport and metabolism; Q, secondary metabolite biosynthesis, transport, and catabolism; R, general function prediction only; S, function unknown; T, signal transduction mechanisms; V, defense mechanisms.

TABLE 2. Induction of DNA repair genes in MR-1 following IR irradiation

Gene ID	Gene name	COG annotation	Relative gene expression or *P* value[a]					
			T5	*P*5	T20	*P*20	T60	*P*60
SO0393	*fis*	Factor for inversion stimulation, Fis: transcriptional activator	1.30	0.09	2.12	0.05	2.58	0.06
SO0690		Site-specific DNA methylase	0.93	0.20	0.91	0.16	5.01	0.04
SO1114	*dinP*	Nucleotidyltransferase/DNA polymerase involved in DNA repair	5.30	0.01	10.05	0.00	7.44	0.00
SO1758		Predicted transcriptional regulator	6.40	0.00	7.60	0.00	5.04	0.00
SO1819	*dinG*	Rad3-related DNA helicases	3.29	0.00	3.42	0.00	2.73	0.01
SO1820	*polB*	DNA polymerase elongation subunit (family B)	3.41	0.02	4.77	0.00	2.93	0.06
SO3061	*topB*	Topoisomerase III	3.56	0.02	7.78	0.00	7.31	0.00
SO3430	*recA*	RecA/RadA recombinase	6.99	0.00	8.98	0.00	6.82	0.01
SO3462	*recN*	ATPase involved in DNA repair	11.77	0.00	13.82	0.00	12.80	0.00
SO3917	*dnaB*	Replicative DNA helicase	0.94	0.46	1.08	0.71	2.63	0.02
SO4364	*recG*	RecG-like helicase	1.82	0.06	2.47	0.03	2.39	0.11
SO4603	*lexA*	SOS-response transcriptional repressors	11.23	0.00	12.89	0.00	11.60	0.00
SO4669	*polA*	DNA polymerase I, 3′→5′ exonuclease and polymerase domains	1.06	0.63	1.26	0.20	3.37	0.01
SOA0012	*umuC*	Nucleotidyltransferase/DNA polymerase involved in DNA repair	3.78	0.05	8.37	0.00	8.42	0.00
SOA0013	*umuD*	SOS-response transcriptional repressors	3.68	0.14	14.58	0.00	16.19	0.00

[a] T5, T20, and T60, relative gene expression at 5, 20, and 60 min, respectively. *P*5, *P*20, and *P*60, *P* values at 5, 20, and 60 min, respectively.

MR-1 prophages. Our previous study demonstrated that following both UVB and UVC irradiation, a large amount of phage particles was released into the medium (23). More phage genes were induced throughout the course of the 1-h recovery period following IR (Table 1) than by UVC or UVB and were induced in a functionally synchronized manner, which is consistent with their functional activation. Thus, it is reasonable to infer that similar to UVR, IR can induce the lytic cycle of prophage in MR-1, which may contribute to this organism's high IR sensitivity.

Induction of DNA damage repair genes. About 2.8% of the MR-1 genome is implicated in DNA replication, recombination, and repair, which is comparable to those of *E. coli* (2.7%) (2) and *D. radiodurans* (3.1%) (19, 29). Our previous studies indicated that photoreactivation, nucleotide excision repair, and the SOS response including mutagenic repair are functional in MR-1 (22). Both RecBCD and RecF recombinational repair pathways are present in MR-1. In addition, MR-1 carries a complete methyl-directed mismatch repair pathway (*mutS*, *mutL*, and *mutH*) and genes (*mutM*, *mutY*, and *mutT*) that are important in preventing mutation due to the oxidized base 8-oxoguanine (12). In contrast, DNA repair systems identified in *D. radiodurans* appear less complex and diverse than those reported for *E. coli* or *S. oneidensis* (4).

We observed a strong SOS response during the recovery period, including the induction of *lexA*, *recA*, *recN*, *dinP*, *dinG*, *polB*, and the *umuDC* operon (Table 2). The induction of most genes lasted for 60 min (Table 2). Furthermore, we observed the induction of several genes that are involved in DNA replication, such as *dnaB*, *polA*, *topB*, and SO0690, which encodes a site-specific DNA methylase, and genes involved in DNA repair, such as *recG* and *fis* (Table 2). Fis is a site-specific DNA binding protein that can bend DNA and facilitates site-specific recombinations (7, 15). In spite of the relatively minor DNA damage inflicted in MR-1 by 40 Gy (4), the SOS response and recombinational repair pathways appear to be highly induced following IR.

Induction of oxidative stress-related genes. MR-1 carries genes encoding catalases, an Fe-containing superoxide dismutase, glutathione peroxidase, glutathione *S*-transferase, organic hydroperoxide resistance protein, and a Dps protein, which protects DNA from oxidative damage (13). In addition, the MR-1 genome encodes ferritin, an iron [Fe(III)] storage protein which also serves as an antioxidant by removing unbound intracellular "free" iron (10). Regarding regulatory genes, it is noteworthy that *S. oneidensis* contains genes that encode 52 proteins related to the LysR family (e.g., SO1328), which includes OxyR, compared to 2 proteins in *D. radiodurans* (10, 19). OxyR is a global transcriptional regulator involved in activating genes that fight oxidative stress.

Several genes that are directly involved in scavenging ROS were strongly up-regulated in MR-1 at 5 min of the 1-h recovery period following IR, which included *katB* (14.9-fold), *ahpC* (11.3-fold), *ahpF* (4.1-fold), *dps* (3.1-fold), and *ccpA* (3.9-fold). *katG-1* also showed a great induction at 5 min (6.4-fold), although the variation among the replicates was high ($P = 0.058$) (Table 3). A slight induction for *trxC* (2.4-fold), which encodes thioredoxin, was observed at 60 min (Table 3). Thioredoxin is a ubiquitous protein known to protect cells against oxidative stress in bacteria (3) as well as in humans (5). In addition, several genes that are involved in degradation, such as *clpB*, *hslV*, and *hslU*, were up-regulated at 60 min (Table 3). Collectively, these results support the finding that irradiated *S. oneidensis* MR-1 mounts a major response to oxidative stress.

Comparison of IR-induced responses between *S. oneidensis* and *D. radiodurans*. Since *D. radiodurans* represents extreme bacterial resistance to IR and is the only other bacterium currently being subjected to transcriptome analyses of its response to IR, we compared transcriptional profiles of MR-1 and *D. radiodurans* to infer which factors might determine radiation resistance and sensitivity in bacteria. A prominent feature of the transcriptome response of *D. radiodurans* recovering from IR is the differential regulation of its TCA cycle, where the isocitrate-to-fumarate steps of the TCA cycle were repressed and the glyoxylate bypass genes (*aceA* and *aceB*) were strongly induced immediately after irradiation (18). Isocitrate lyase (AceA) converts isocitrate into succinate and glyoxylate, allowing carbon that entered the TCA cycle to bypass the formation of α-ketoglutarate and succinyl coenzyme A. It has been proposed that such regulation might strongly

TABLE 3. Induction of genes in defending against oxidative stress in MR-1 following IR irradiation

Gene ID	Gene name	COG annotation	Relative gene expression or TDEST value[a]					
			T5	*P*5	T20	*P*20	T60	*P*60
SO0956	*ahpF*	Alkyl hydroperoxide reductase, large subunit	4.13	0.01	1.14	0.68	1.07	0.84
SO0958	*ahpC*	Peroxiredoxin	11.25	0.00	1.97	0.09	1.07	0.98
SO3577	*clpB*	ATPase with chaperone activity, ATP-binding subunit	1.16	0.44	0.96	0.78	2.97	0.01
SO4162	*hslV*	ATP-dependent protease HslVU (ClpYQ), peptidase subunit	1.06	0.79	1.06	0.84	4.06	0.03
SO4163	*hslU*	ATP-dependent protease HslVU (ClpYQ), ATPase subunit	0.95	0.60	1.11	0.66	3.42	0.01
SO4697	*gst*	Glutathione *S*-transferase	0.92	0.74	1.30	0.39	3.68	0.01
SO0452	*trxC*	Thioredoxin	2.08	0.10	1.75	0.01	2.41	0.01
SO0760	*amt*	Ammonia permease	1.45	0.77	2.03	0.00	1.03	0.96
SO1070	*katB*	Catalase	14.89	0.02	1.62	0.92	1.44	0.86
SO1145	*mgtE-1*	Mg/Co/Ni transporter MgtE (contains CBS domain)	0.96	0.85	1.56	0.30	5.80	0.04
SO1158		DNA-binding ferritin-like protein (oxidative damage protectant)	3.09	0.01	1.28	0.49	1.07	0.93
SO2178	*ccpA*	Cytochrome *c* peroxidase	3.94	0.01	1.29	0.33	0.86	0.35
SO4423.1		Outer membrane receptor proteins, related to Fe transport	1.28	0.29	2.74	0.04	2.70	0.18
SO2881	*sodB*	Superoxide dismutase, Fe	1.51	0.04	0.91	0.69	0.66	0.37

[a] T5, T20, and T60, relative gene expression at 5, 20, and 60 min, respectively. *P*5, *P*20, and *P*60, *P* values at 5, 20, and 60 min, respectively.

suppress oxidative stress in *D. radiodurans* during recovery, perhaps as a mechanism to prevent additional loss of genome integrity (10, 18). This contrasts with the response we observed for *S. oneidensis* recovering from IR, which showed no significant change in TCA cycle expression, including its glyoxylate bypass. Furthermore, an early induction of catalase was observed in *S. oneidensis* (Table 3), whereas there was no change in expression of *kat* in *D. radiodurans* (18). It appeared that different strategies were used in *D. radiodurans* and *S. oneidensis* to fight against oxidative stress following IR, perhaps as a result of higher levels of oxidative stress generated in *S. oneidensis* during irradiation and recovery.

Our approach to determining the expression dynamics of MR-1 recovering from IR involved growth to the logarithmic phase and irradiation (40 Gy) on ice, followed by recovery in the same medium at 30°C. Whether differences in growth medium (LB versus Davis), radiation dose (15 kGy versus 40 Gy), and recovery medium (fresh medium versus the same medium) contribute to the difference in transcriptome between *D. radiodurans* and *S. oneidensis* needs further evaluation.

Conclusion. Approximately 80% of *S. oneidensis* cells are killed following exposure to 40 Gy, which causes less than 1 DSB per genome (5.1 Mbp) and about 40 SSB per genome (4, 17). In light of the strong induction of DNA repair and protection systems elicited in irradiated *S. oneidensis*, the relatively minor DNA damage does not explain the high levels of cell killing. As a respiratory generalist, MR-1 is rich in iron-containing proteins but low in Mn. Accumulation of intracellular Mn(II) in *D. radiodurans* has been implicated in recovery from IR by scavenging $O_2^{\cdot -}$ and preventing decomposition of H_2O_2 to $HO^{\cdot}$ (4, 10). In contrast, a sudden increase of "free" iron due to protein damage would cause a proliferation of the ROS in MR-1 during and following IR, which may predispose *S. oneidensis* cells to a burst of oxidative stress at the onset of recovery. This, coupled with the induction of lytic phages suggested by the expression data, likely explains the high sensitivity of MR-1 to IR.

Microarray data accession number. The microarray data have been deposited in the Gene Expression Omnibus database under accession no. GSE3876.

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