CHAPTER 10.5

Regulators of oxidative stress response genes in *Escherichia coli* and their conservation in bacteria

Herb E. Schellhorn, Mohammad Mohiuddin, Sarah M. Hammond, and Steven Botts
Department of Biology, McMaster University, Hamilton, Ontario, Canada

10.5.1 Introduction

Oxidative stress in the bacterial cell, caused by endogenous metabolism or encountered from exogenous sources, elicits a complex response that can be divided into two distinct components. The preventative component consists of enzymes that mitigate oxidative stress by converting the reactive species into less toxic intermediates through reduction or dismutation. The bacterial cell also has a battery of enzymes that can repair damage caused by oxidative stress. In this review, we will focus on recent findings on oxidative stress regulators to understand how the preventative component has arisen through evolutionary processes.

Characterization of oxidative stress effects in bacterial cells has been greatly aided by the use of genomic technologies (including microarray and proteomics) that have facilitated the identification of many new, previously unknown components of the protective response. Technologies that enable probing events at the single-cell level have also contributed to our understanding since bacteria exist as populations of cells and even small numbers of cells can contribute enormously to the subsequent outgrowth of populations. Bacterial oxidative stress–mediated damage, although historically studied at the population level using enzyme assays performed on culture samples, can now be probed and individual cells can be studied using microscopic and fluorometric-based assays that provide information on the status of single cells. These data have shown that oxidative stress does not affect all cells uniformly and that, in fact, heterogeneity in aging cultures is the hallmark of bacterial growth that is undoubtedly important in survival from toxic levels of stress. This review will focus on the role of oxidative stress responses in *E. coli* as a model with comparisons to key bacterial groups that share regulatory homologs.

10.5.2 Levels of bacterial catalase and superoxide dismutase are closely linked to the flux of reactive oxygen species in the cell

Reactive oxygen–mediated stress in the bacterial cell may be regarded as episodic in nature, depending on the mode of metabolism of the cell (respiratory versus fermentative) or the presence of exogenous sources of reactive oxygen species (ROS) (Figure 10.5.1). Regulators directly sense the presence of ROS through oxidation and reduction of specific amino acid residues.

Superoxide dismutase is produced in response to both high oxygen tension and excess iron. MnSOD is induced by SoxRS (Pomposiello and Demple, 2000), and in *Salmonella*, MnSOD together with FeSOD are activated by the Fur protein through small RNAs, *rfrA* and *rfaB* (Troxell *et al.*, 2011). The CuZnSOD-encoding *sodC* gene is repressed by the Fnr regulator and thus is not expressed under anaerobic conditions (Gort *et al.*, 1999) when superoxide is absent. Both peroxidase enzymes in *E. coli* are part of the OxyR regulon and are thus expressed during increased peroxide flux (Tartaglia *et al.*, 1989). Both CuZnSOD (Gort *et al.*, 1999) and the HPII catalase (Schellhorn and Hassan, 1988) increase 20–30-fold during growth to the stationary phase, allowing the cell to adapt to a slow growth state where specific *de novo* induction may be less feasible. *E. coli* peroxidases are controlled by OxyR (Tartaglia *et al.*, 1989) and are induced by low concentrations of hydrogen peroxide.

10.5.3 Oxidative stress overlaps with other stress responses, including the RpoS regulon

During the exponential phase, the cell can rapidly adjust to changes in the flux of ROS by induction of the appropriate

Stress and Environmental Regulation of Gene Expression and Adaptation in Bacteria, First Edition. Edited by Frans J. de Bruijn.

	Reaction			Gene	Protein	Regulators
Equation 1	$2O_2^{\cdot -} + 2H^+$	*Superoxide dismutase* →	$H_2O_2 + O_2$	*sodA*	MnSOD	Fur (+), SoxRS (+)
				sodB	FeSOD	Fur (+)
				sodC	CuZnSOD	RpoS (+), Fnr (+)
Equation 2	$2H_2O_2$	*Catalase* →	$2H_2O + O_2$	*katE*	HPII	RpoS (+)
				katG	HPI	OxyR (+)
Equation 3	$H_2O_2 + AH_2$	*Peroxidase* →	$2H_2O + A$	*ahpCH*	AHP	OxyR (+)
				katG	HPI	OxyR (+)

Figure 10.5.1 Regulation of oxidative stress enzymes in *Escherichia coli*. "A" is a cellular reductant.

enzymes. However, in the stationary phase, *de novo* protein synthesis is substantially reduced; yet, at the high cell densities during this phase of growth, oxidative stress can be elevated due to increased cell density and increased respiratory activity as cells use carbon sources that require respiratory metabolism. Under such conditions, rapid adaptation mechanisms such as those controlled by OxyR and SoxRS are likely less effective. However, in *E. coli*, during adaptation to the stationary phase, many genes are controlled by the second vegetative sigma factor, RpoS, which controls many diverse functions. RpoS is a general stress response regulator that controls hundreds of genes required for adaptation. Consistent with this physiological role, catalases (e.g., KatE–HPII) are important in eliminating hydrogen peroxide levels at high concentrations, whereas peroxidases (including AphCF and HPI) are critical at lower concentrations (Imlay, 2013).

The *rpoS* gene is found in proteobacteria, primarily in the gamma-, beta-, and deltaproteobacteria (Chiang and Schellhorn, 2010), having arisen through a duplication of the *rpoD* gene encoding the main vegetative sigma factor, sigma 70. Though controlling many functions, not surprisingly, some oxidative stress genes are specifically expressed in the stationary phase. Both catalase (HPII) and superoxide dismutase (SodC) enzymes are highly RpoS dependent and thus are part of the protective regulon that ensures cell survival during periods of slow growth. Examination of RpoS regulon composition between *Pseudomonas aeruginosa* and *E. coli* indicates that relatively few members are conserved between these two proteobacteria. The RpoS regulon may therefore represent a reconfigurable system that allows its incorporation in expression of protective functions that are needed for a particular or specific bacterial niche (Schellhorn, 2014). Proteobacteria constitute a large group of bacteria that possess substantial metabolic diversity, and they include many pathogens that may experience oxidative stress within the host or as free-living bacteria. Conserved control of components of the oxidative stress response by the RpoS regulator allows the bacterium to express high levels of relevant protective activity precisely calibrated to its immediate metabolic needs.

10.5.4 Evolutionary conservation of the OxyR peroxide regulator

OxyR, a member of the LysR family of transcriptional regulators, serves as a bacterial primary defense mechanism against hydrogen peroxide stress (Jang and Imlay, 2010; Zheng *et al.*, 2001; see Chapters 10.1 and 10.2). Characteristic of the LysR protein family, OxyR negatively regulates its own expression but positively regulates *oxyS*, an adjacent small RNA that integrates the peroxide stress response with the cellular stress response (Li and He, 2012).

In response to elevated levels of hydrogen peroxide (intracellular or extracellular), OxyR, in *E. coli*, activates the transcription of 28 dependent promoters as reviewed in Chiang and Schellhorn (2012). This induction depends on the oxidation state of OxyR and cooperative binding of oxidized OxyR with RNA polymerase (Lee *et al.*, 2004). The cooperatively bound RNA polymerase then positively regulates transcription (Lee *et al.*, 2004; Tao *et al.*, 1995). Reduced OxyR is a negative regulator of genes (e.g., *agn43*, *stiA*, and *pntAA*) in some species, including *Salmonella enterica* and *P. aeruginosa* (Henderson and Owen, 1999; Seymour *et al.*, 1996; Wei *et al.*, 2012). The OxyR regulon, as part of the response against oxidative stress, includes *gorA* (glutathione reductase) and *grxA* (glutaredoxin). The glutaredoxin–glutathione system itself comprises a feedback control mechanism for OxyR, slowly reducing oxidized OxyR to the fully reduced form (Garcia-Santamarina *et al.*, 2014). In addition to peroxide resistance, OxyR also protects against heat stress (Wei *et al.*, 2012), singlet oxygen (Kim *et al.*, 2002), lipid peroxidation–mediated oxidative stress (Daugherty *et al.*, 2012; Yoon *et al.*, 2002), and phagocyte ingestion (Staudinger *et al.*, 2002).

OxyR regulon homologs are present in the Proteobacteria, Actinobacteria, and Bacteroidetes phyla, as reviewed in Chiang and Schellhorn (2012). Among the five classes of Proteobacteria, OxyR homologs are present in the most recently diverged gammaproteobacteria, in betaproteobacteria, and in alphaproteobacteria and are absent in delta- and epsilonproteobacteria (Rodionov *et al.*, 2004; van Vliet *et al.*, 1999). Although

functional regulation by OxyR is conserved in most gammaproteobacteria, one exception (at least) is *Legionella pneumophila*. In *L. pneumophila*, the OxyR regulon, denoted as $OxyR_{Lp}$, instead of responding to oxidative stress, regulates the transition of this bacterial species from a vegetative state to a resilient cyst-like transmissible form (LeBlanc *et al.*, 2008). The composition and size of the OxyR regulon vary among classes of Proteobacteria (Chiang and Schellhorn, 2010). OxyR provides protection against exposure to atmospheric oxygen in the anaerobic phylum Bacteroidetes (Diaz *et al.*, 2006). *Bacteroidetes fragilis* OxyR, with 40% identity to aerobic bacteria, regulates an acute response regulon of 13 genes to minimize the impact of ROS to maintain intracellular redox balance (Sund *et al.*, 2008). In Actinobacteria, OxyR also confers protection against hydrogen peroxide. For instance, in *Streptomyces* spp. and in *Mycobacterium marinum*, overexpression of OxyR confers hydrogen peroxide resistance through induction of *ahpCD* and *ahpC* expression, respectively (Hahn *et al.*, 2002; Pagan-Ramos *et al.*, 2006).

10.5.5 Control of superoxide responses by SoxRS

The adjacent and divergently transcribed genes *soxR* and *soxS* comprise a two-component regulatory system for mediating oxidative stress in *E. coli*. First identified in 1991, the SoxRS regulon is composed of more than 100 genes (Blanchard *et al.*, 2007; Wu and Weiss, 1991). In *E. coli*, the activation of SoxR (superoxide response) induces expression of *soxS*, which regulates key genes involved in the superoxide radical response (Nunoshiba *et al.*, 1992).

SoxR regulates resistance to oxidative stress in *E. coli* following exposure to the superoxide inducer paraquat (Greenberg *et al.*, 1990), nitric oxide (Nunoshiba *et al.*, 1993), and hydrogen peroxide (Manchado *et al.*, 2000). In addition to these compounds, SoxR is also directly activated by redox-cycling drugs in anaerobic environments (Singh *et al.*, 2013). The oxidation of two (2Fe-2S) clusters in SoxR results in a 100-fold induction of *soxS* (Hidalgo and Owen, 1994). Upon oxidation, SoxR complexes with the spacer region of the *soxS* promoter to facilitate binding of RNA polymerase and downstream transcription (Hidalgo and Owen, 1997). Several SoxRS regulon genes are active in mitigating oxidative stress, including *sodA* (superoxide dismutation) and *nfo* (DNA repair) (Li and Demple, 1994).

Homologs of the SoxR regulator have been found in both proteo- and actinobacteria, although they remain unidentified in Bacteroidetes (Ohara *et al.*, 2006). SoxRS responds to oxidative stress in both *Escherichia* and *Shigella* spp., where *soxRS* mutants show increased sensitivity to superoxide (Daugherty *et al.*, 2012). The SoxRS regulon additionally confers multiple-drug resistance in both *E. coli* and the genera *Salmonella* and *Klebsiella* in gammaproteobacteria (Kehrenberg *et al.*, 2009; Mosel *et al.*, 2013). SoxR homologs in *Vibrio* spp. and *P. aeruginosa*, respectively, share 55 and 62% sequence identity with *E. coli* (Kobayashi and Tagawa, 2004; Vattanaviboon *et al.*, 2003). The SoxRS-mediated superoxide radical response is present in many gammaproteobacteria (*E. coli*, *Shigella flexneri*, *Salmonella* spp., *Klebsiella pneumoniae*, and *Vibrio* spp.) but is absent in *Pseudomonas* spp., where SoxRS regulates drug efflux pump genes (Palma *et al.*, 2005).

10.5.6 Oxidative stress and bacterial persistence

Within homogeneous populations of bacteria exposed to a given antibiotic, a small fraction of cells are nongrowing and therefore refractile to the lethal effects of bactericidal antibiotics. These so-called persister cells (see Chapter 6.3) represent an important type of phenotypic plasticity that allows a population of cells to survive transient exposure to antimicrobial agents or conditions. Whether persister cells arise through stochastic or deterministic processes is an important issue, and although this question is not fully resolved, it is clear that genetic determinants can affect the fraction of persistent cells within a population. Populations of cells deficient in *rpoS* have higher levels of persister cells. This may be due to reduced levels of protective enzymes that result in oxidative stress–mediated inhibition of growth within subpopulations (Wang *et al.*, 2011). Consistent with the idea that inhibition of growth is key to persistence, toxin–antitoxin systems (Dorr *et al.*, 2010; Wang *et al.*, 2011; see Chapter 2.7 and Section 6) have been implicated in the development of persistence. These results suggest that the status of individual cells within a population may be a key to understanding how bacterial populations survive oxidative stress and related antibiotic stress to propagate successfully.

10.5.7 Complexity of oxidative stress responses revealed by transcriptome technologies

Oxidative stress responses reflect an interplay between the cell's physiological adaptation to ROS, intracellular iron availability, and repair factors that maintain viability. Uncovering such complexity requires interrogation of genome-wide responses using transcriptome-based techniques, including microarrays and RNA-seq experiments, under specific conditions such as slow growth or altered expression in pathogenic strains or in relation to other genetic factors such as phosphate regulation (Table 10.5.1). While many oxidative stress–related responses have been defined by comparing expression results of wild-type strains to regulatory deletion mutants, a more nuanced approach, using mutants that robustly model physiologically relevant low-level ROS fluxes, may provide important information. For example, initial characterization of the OxyR response (Zheng *et al.*, 2001) revealed many genes that form the core primary defense against hydrogen peroxide. Later studies extended

Table 10.5.1 Transcriptome-based identification of oxidative responses in bacteria.

Species	Oxidative response functions examined	Main finding	Reference
Escherichia coli	OxyR	Defined the OxyR regulon using deletion mutants.	Zheng *et al.* (2001)
E. coli	PhoB, RpoS	Phosphate limitation alters expression of oxidative stress genes.	Chekabab *et al.* (2014)
E. coli	OxyR, HPX	Used catalase and peroxide mutants to identify new OxyR regulon members.	Mancini *et al.* (2015)
E. coli	RpoS	Environment selection and host selection result in increased nutrient scavenging at the expense of expression of oxidative stress response genes.	Parker *et al.* (2012)
Salmonella enterica serovar Enteritidis	RpoS	Expression of oxidative stress genes is reduced in poorly pathogenic strains versus highly pathogenic strains.	Shah (2014)
Pseudomonas sp.	OxyR	Expression of ferric reductase is critical for oxidative stress–dependent, antibiotic-mediated killing.	Yeom *et al.* (2010)

this, using catalase and peroxide mutants in which exposure levels mimic normal intracellular levels of this oxidant (Mancini and Imlay, 2015) that are known to modulate activity of the OxyR protein (Imlay, 2015). These types of studies reveal that oxidative stress responses are intimately associated with biosynthetic processes, including heme biosynthesis (Mancini and Imlay, 2015).

10.5.8 Concluding remarks

Bacterial responses to oxidative stress, controlled by conserved regulators, are highly adaptable, allowing the cell to increase expression of many metabolic functions that are coordinately controlled to reduce the effects of stress. It is now clear that oxidative stresses overlap with other, related bacterial responses, and considerable future work will be needed to uncover the physiological responses to complex stresses. For example, proline catabolism in *E. coli* appears to produce sufficient oxidative stress to induce the OxyR regulon, indicating that nonrespiratory pathways may contribute to the bacterial stress response (Zhang *et al.*, 2015). Selective killing by oxidative stress results in population bottlenecks, which can provide strong selection of new stress-related survival traits. For this reason, responses in individual cells are likely critical in allowing populations to propagate, and understanding stochastic and deterministic factors at the individual cell level will likely be a focus of future research.

Acknowledgements

We thank members of the Schellhorn laboratory for many helpful discussions. Work in the Schellhorn laboratory is supported by the Natural Sciences and Engineering Research Council (Canada) through its Discovery grants program (Grant no. RGPIN/46644-2010).

References

Blanchard JL, Wholey WY, Conlon EM, Pomposiello PJ. 2007. Rapid changes in gene expression dynamics in response to superoxide reveal SoxRS-dependent and independent transcriptional networks. *PloS One* 2(11): e1186.

Chekabab SM, Jubelin G, Dozois CM, Harel J. 2014. PhoB activates *Escherichia coli* O157:H7 virulence factors in response to inorganic phosphate limitation. *PloS One* 9(4): e94285.

Chiang SM, Schellhorn HE. 2010. Evolution of the RpoS regulon: origin of RpoS and the conservation of RpoS-dependent regulation in bacteria. *Journal of Molecular Evolution* 70(6): 557–571.

Chiang SM, Schellhorn HE. 2012. Regulators of oxidative stress response genes in *Escherichia coli* and their functional conservation in bacteria. *Archives of Biochemistry and Biophysics* 525(2): 161–169.

Daugherty A, Suvarnapunya AE, Runyen-Janecky L. 2012. The role of *oxyR* and *soxRS* in oxidative stress survival in *Shigella flexneri*. *Microbiological Research* 167(4): 238–245.

Diaz PI, Slakeski N, Reynolds EC, Morona R, Rogers AH, *et al.* 2006. Role of *oxyR* in the oral anaerobe *Porphyromonas gingivalis*. *Journal of Bacteriology* 188(7): 2454–2462.

Dorr T, Vulic M, Lewis K. 2010. Ciprofloxacin causes persister formation by inducing the TisB toxin in *Escherichia coli*. *PLoS Biology* 8(2).

Garcia-Santamarina S, Boronat S, Hidalgo E. 2014. Reversible cysteine oxidation in hydrogen peroxide sensing and signal transduction. *Biochemistry* 53(16): 2560–2580.

Gort AS, Ferber DM, Imlay JA. 1999. The regulation and role of the periplasmic copper, zinc superoxide dismutase of *Escherichia coli*. *Molecular Microbiology* 32(1): 179–191.

Greenberg JT, Monach P, Chou JH, Josephy PD, Demple B. 1990. Positive control of a global antioxidant defense regulon activated by superoxide-generating agents in *Escherichia coli*. *Proceedings of the National Academy of Sciences of the USA* 87(16): 6181–6185.

Hahn JS, Oh SY, Roe JH. 2002. Role of OxyR as a peroxide-sensing positive regulator in *Streptomyces coelicolor* A3(2). *Journal of Bacteriology* 184(19): 5214–5222.

Henderson IR, Owen P. 1999. The major phase-variable outer membrane protein of *Escherichia coli* structurally resembles the

immunoglobulin A1 protease class of exported protein and is regulated by a novel mechanism involving *dam* and *oxyR*. *Journal of Bacteriology* 181(7): 2132–2141.

Hidalgo E, Demple B. 1994. An iron-sulfur center essential for transcriptional activation by the redox-sensing SoxR protein. *EMBO Journal* 13(1): 138–146.

Hidalgo E, Demple B. 1997. Spacing of promoter elements regulates the basal expression of the *soxS* gene and converts SoxR from a transcriptional activator into a repressor. *EMBO Journal* 16(5): 1056–1065.

Imlay JA. 2013. The molecular mechanisms and physiological consequences of oxidative stress: lessons from a model bacterium. *Nature Reviews Microbiology* 11(7): 443–454.

Imlay JA. 2015. Diagnosing oxidative stress in bacteria: not as easy as you might think. *Current Opinion in Microbiology* 24C: 124–131.

Jang S, Imlay JA. 2010. Hydrogen peroxide inactivates the *Escherichia coli* Isc iron-sulphur assembly system, and OxyR induces the Suf system to compensate. *Molecular Microbiology* 78(6): 1448–1467.

Kehrenberg C, Cloeckaert A, Klein G, Schwarz S. 2009. Decreased fluoroquinolone susceptibility in mutants of *Salmonella* serovars other than Typhimurium: detection of novel mutations involved in modulated expression of *ramA* and *soxS*. *Journal of Antimicrobial Chemotherapy* 64(6): 1175–1180.

Kim SY, Kim EJ, Park JW. 2002. Control of singlet oxygen-induced oxidative damage in *Escherichia coli*. *Journal of Biochemistry and Molecular Biology* 35(4): 353–357.

Kobayashi K, Tagawa S. 2004. Activation of SoxR-dependent transcription in *Pseudomonas aeruginosa*. *Journal of Biochemistry* 136(5): 607–615.

LeBlanc JJ, Brassinga AK, Ewann F, Davidson RJ, Hoffman PS. 2008. An ortholog of OxyR in *Legionella pneumophila* is expressed postexponentially and negatively regulates the alkyl hydroperoxide reductase (*ahpC2D*) operon. *Journal of Bacteriology* 190(10): 3444–3455.

Lee C, Lee SM, Mukhopadhyay P, Kim SJ, Lee SC, *et al.* 2004. Redox regulation of OxyR requires specific disulfide bond formation involving a rapid kinetic reaction path. *Nature Structural & Molecular Biology* 11(12): 1179–1185.

Li Y, He ZG. 2012. The mycobacterial LysR-type regulator OxyS responds to oxidative stress and negatively regulates expression of the catalase-peroxidase gene. *PloS One* 7(1): e30186.

Li Z, Demple B. 1994. SoxS, an activator of superoxide stress genes in *Escherichia coli*. Purification and interaction with DNA. *Journal of Biological Chemistry* 269(28): 18371–18377.

Manchado M, Michan C, Pueyo C. 2000. Hydrogen peroxide activates the SoxRS regulon in vivo. *Journal of Bacteriology* 182(23): 6842–6844.

Mancini S, Imlay JA. 2015. The induction of two biosynthetic enzymes helps *Escherichia coli* sustain heme synthesis and activate catalase during hydrogen peroxide stress. *Molecular Microbiology*.

Mosel M, Li L, Drlica K, Zhao X. 2013. Superoxide-mediated protection of *Escherichia coli* from antimicrobials. *Antimicrobial Agents & Chemotherapy* 57(11): 5755–5759.

Nunoshiba T, deRojas-Walker T, Wishnok JS, Tannenbaum SR, Demple B. 1993. Activation by nitric oxide of an oxidative-stress response that defends *Escherichia coli* against activated macrophages. *Proceedings of the National Academy of Sciences of the USA* 90(21): 9993–9997.

Nunoshiba T, Hidalgo E, Amabile Cuevas CF, Demple B. 1992. Two-stage control of an oxidative stress regulon: the *Escherichia coli* SoxR protein triggers redox-inducible expression of the *soxS* regulatory gene. *Journal of Bacteriology* 174(19): 6054–6060.

Ohara N, Kikuchi Y, Shoji M, Naito M, Nakayama K. 2006. Superoxide dismutase-encoding gene of the obligate anaerobe *Porphyromonas gingivalis* is regulated by the redox-sensing transcription activator OxyR. *Microbiology* 152(Pt 4): 955–966.

Pagan-Ramos E, Master SS, Pritchett CL, Reimschuessel R, Trucksis M, *et al.* 2006. Molecular and physiological effects of mycobacterial *oxyR* inactivation. *Journal of Bacteriology* 188(7): 2674–2680.

Palma M, Zurita J, Ferreras JA, Worgall S, Larone DH, *et al.* 2005. *Pseudomonas aeruginosa* SoxR does not conform to the archetypal paradigm for SoxR-dependent regulation of the bacterial oxidative stress adaptive response. *Infection and Immunity* 73(5): 2958–2966.

Parker CT, Kyle JL, Huynh S, Carter MQ, Brandl MT, *et al.* 2012. Distinct transcriptional profiles and phenotypes exhibited by *Escherichia coli* O157:H7 isolates related to the 2006 spinach-associated outbreak. *Applied Environmental Microbiology* 78(2): 455–463.

Pomposiello PJ, Demple B. 2000. Identification of SoxS-regulated genes in *Salmonella enterica* serovar Typhimurium. *Journal of Bacteriology* 182(1): 23–29.

Rodionov DA, Dubchak I, Arkin A, Alm E, Gelfand MS. 2004. Reconstruction of regulatory and metabolic pathways in metal-reducing delta-proteobacteria. *Genome Biology* 5(11): R90.

Schellhorn HE. 2014. Elucidating the function of the RpoS regulon. *Future Microbiology* 9(4): 497–507.

Schellhorn HE, Hassan HM. 1988. Transcriptional regulation of *katE* in *Escherichia coli* K-12. *Journal of Bacteriology* 170(9): 4286–4292.

Seymour RL, Mishra PV, Khan MA, Spector MP. 1996. Essential roles of core starvation-stress response loci in carbon-starvation-inducible cross-resistance and hydrogen peroxide-inducible adaptive resistance to oxidative challenge in *Salmonella typhimurium*. *Molecular Microbiology* 20(3): 497–505.

Shah DH. 2014. RNA sequencing reveals differences between the global transcriptomes of *Salmonella enterica* serovar enteritidis strains with high and low pathogenicities. *Applied and Environmental Microbiology* 80(3): 896–906.

Singh AK, Shin JH, Lee KL, Imlay JA, Roe JH. 2013. Comparative study of SoxR activation by redox-active compounds. *Molecular Microbiology* 90(5): 983–996.

Staudinger BJ, Oberdoerster MA, Lewis PJ, Rosen H. 2002. mRNA expression profiles for *Escherichia coli* ingested by normal and phagocyte oxidase-deficient human neutrophils. *Journal of Clinical Investigation* 110(8): 1151–1163.

Sund CJ, Rocha ER, Tzianabos AO, Wells WG, Gee JM, *et al.* 2008. The *Bacteroides fragilis* transcriptome response to oxygen and H_2O_2: the role of OxyR and its effect on survival and virulence. *Molecular Microbiology* 67(1): 129–142.

Tao K, Zou C, Fujita N, Ishihama A. 1995. Mapping of the OxyR protein contact site in the C-terminal region of RNA polymerase alpha subunit. *Journal of Bacteriology* 177(23): 6740–6744.

Tartaglia LA, Storz G, Ames BN. 1989. Identification and molecular analysis of *oxyR*-regulated promoters important for the bacterial adaptation to oxidative stress. *Journal of Molecular Biology* 210(4): 709–719.

Troxell B, Fink RC, Porwollik S, McClelland M, Hassan HM. 2011. The Fur regulon in anaerobically grown *Salmonella enterica* sv. Typhimurium: identification of new Fur targets. *BMC Microbiology* 11: 236.

van Vliet AH, Baillon ML, Penn CW, Ketley JM. 1999. *Campylobacter jejuni* contains two Fur homologs: characterization of iron-responsive regulation of peroxide stress defense genes by the PerR repressor. *Journal of Bacteriology* 181(20): 6371–6376.

Vattanaviboon P, Panmanee W, Mongkolsuk S. 2003. Induction of peroxide and superoxide protective enzymes and physiological cross-protection against peroxide killing by a superoxide generator in *Vibrio harveyi*. *FEMS Microbiology Letters* 221(1): 89–95.

Wang XX, Kim Y, Hong SH, Ma Q, Brown BL, *et al.* 2011. Antitoxin MqsA helps mediate the bacterial general stress response. *Nature Chemical Biology* 7(6): 359–366.

Wei Q, Minh PN, Dotsch A, Hildebrand F, Panmanee W, *et al.* 2012. Global regulation of gene expression by OxyR in an important human opportunistic pathogen. *Nucleic Acids Research* 40(10): 4320–4333.

Wu J, Weiss B. 1991. Two divergently transcribed genes, *soxR* and *soxS*, control a superoxide response regulon of *Escherichia coli*. *Journal of Bacteriology* 173(9): 2864–2871.

Yeom J, Imlay JA, Park W. 2010. Iron homeostasis affects antibiotic-mediated cell death in *Pseudomonas* species. *Journal of Biological Chemistry* 285(29): 22689–22695.

Yoon SJ, Park JE, Yang JH, Park JW. 2002. OxyR regulon controls lipid peroxidation-mediated oxidative stress in *Escherichia coli*. *Journal of Biochemistry and Molecular Biology* 35(3): 297–301.

Zhang L, Alfano JR, Becker DF. 2015. Proline metabolism increases *katG* expression and oxidative stress resistance in *Escherichia coli*. *Journal of Bacteriology* 197(3): 431–440.

Zheng M, Wang X, Templeton LJ, Smulski DR, LaRossa RA, *et al.* 2001. DNA microarray-mediated transcriptional profiling of the *Escherichia coli* response to hydrogen peroxide. *Journal of Bacteriology* 183(15): 4562–4570.