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ABSTRACT: The Role of the Novel Diguanylate Cyclase PG_0686 in Oxidative Stress Resistance in Porphyromonas gingivalis W83

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by

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ABSTRACT OF THE DISSERTATION

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The survival/ adaptation of *Porphyromonas gingivalis* to the inflammatory environment of the periodontal pocket requires an ability to overcome oxidative stress. Several functional classes of genes, depending on the severity and duration of the exposure, were induced in *P. gingivalis* under H_2O_2-induced oxidative stress, including the *PG0686* gene which was upregulated ca. 10-fold. In addition, its upregulation was also observed in the presence of oxygen and nitric oxide. This study is aimed to further characterize the function of this gene in response to H_2O_2. PG_0686, annotated as a hypothetical protein of unknown function, is a 60 kDa protein with a diguanylate cyclase (DGC)-like fold and carries other domains including hemerythrin, a PAS10 domain, and Domain of Unknown Function (DUF)-1858. PG_0686 is missing the classical active site conserved sequence motif (GGD(/E)EF) commonly observed in the DGC of other bacteria. PG_0686-related proteins are observed in other anaerobic bacterial species. The isogenic mutant *P. gingivalis* FLL361 (Δ*PG0686::ermF*) showed increased sensitivity to H_2O_2 and decreased gingipain activity compared to the parent strain. Transcriptome analysis of *P. gingivalis* FLL361 showed the dysregulation of several gene clusters/operons, known oxidative stress resistance genes and

transcriptional regulators including PG_2212, CdhR and PG_1181 that were upregulated under normal anaerobic conditions. The purified recombinant PG_0686 protein can catalyze c-di-GMP formation from GTP. The intracellular level of c-di-GMP in *P. gingivalis* FLL361 was significantly decreased compared to the parent strain. Collectively, our data suggest a global regulatory property for PG_0686 that may be part of an unconventional second messenger signaling system in *P. gingivalis*. Moreover, it may coordinately regulate a pathway(s) vital for protection against environmental stress and is significant in the pathogenicity of the *P. gingivalis*, and likely other anaerobes.