

***Porphyromonas gingivalis* Outer Membrane Vesicles Modulate Biofilm Morphology**

Joshua Ortiz, Alisa King, Angel Reddy, Andrea Gonzalez, Steve Coats, Sumita Jain, and Sarah Alaei

The biofilm forming bacterium *Porphyromonas gingivalis* (*Pg*) produces a large amount of Outer Membrane Vesicles (OMVs), transport vessels for biomolecules such as proteases. As a keystone pathogen in chronic periodontitis, *Pg* is able to manipulate the host's innate immune system and alter neighboring commensal bacteria, resulting in dysbiosis of the microbiota. The *Pg* strains 33277 and 381 are genetically similar but differ in the abundance of OMVs produced with 381 producing significantly more. We therefore investigated whether this difference in OMV abundance relates to any biofilm morphology variance between the two strains. *Pg* 33277, 381, and isogenic mutants of each with lipid A modifying enzyme gene deletions ($\Delta lpxE$, $\Delta lpxF$, $\Delta lpxZ$) were cultured for biofilm assays, quantification of OMV production, and TEM image comparison. Results reveal that 381 generates significantly more OMVs when compared to 33277. Analysis of mutants showed reduction of OMV production in 33277 $\Delta lpxF$ that correlated with increased biofilm density compared to its wild type. Biofilms of 381 were also much denser than 33277, with cells congregating in columnar clusters. OMVs contributed to biofilm dispersal based on their abundance. OMVs of 381 possessed lower protease activity when compared to the protease activity of 33277. This suggests that there may be strain specific differences in cargo transported by OMVs. These findings may help describe the underlying role *Pg* OMVs have in the pathogenesis of chronic periodontitis.