

## INTERACTIONS OF HUMAN UMBILICAL VEIN ENDOTHELIAL CELLS WITH TOBACCO TREATED *STREPTOCOCCUS MUTANS*

**Branden Lanier**<sup>1</sup>, L. Jack Windsor<sup>1</sup>, and Richard L. Gregory<sup>1</sup>,

<sup>1</sup>Department of Oral Biology, Indiana University School of Dentistry, Indianapolis, IN 46202

*Streptococcus mutans* and tobacco are risk factors for atherosclerosis. The objective of this study was to determine the ability that a spaP isogenic defective mutant of *S. mutans* UA 159 has on binding to Human Umbilical Vein Endothelial Cells (HUVEC) when treated with tobacco products and what second messenger signals are involved. The study was conducted to examine the effects that various concentrations of cigarette smoke condensate (CSC)- and nicotine have on *S. mutans* cell cytotoxicity and expression of cytokines and growth factors from HUVECs. *S. mutans* was grown at 37°C and planktonic and biofilm cells were separated from the culture supernatant. The supernatant was discarded the cells were washed, sterilized with formaldehyde and washed again to remove the formaldehyde. The concentrations of the various *S. mutans* cells were standardized to the same concentration (absorbance of  $0.50 \pm 0.01$ ) by spectroscopy at a wavelength of 600 nm. The lowest non-toxic levels of the sterilized bacterial cells were used to treat HUVECs for 72 hours and cytotoxicity was determined by lactate dehydrogenase (LDH) assays. The cytokine/growth factor expression will be determined by antibody protein arrays. The results are expected to indicate an increase in cytotoxicity with increasing cell concentrations, along with increased pro-inflammatory cytokine/growth factors expression by the HUVECs treated with tobacco treated *S. mutans* compared to *S. mutans* that was not treated with tobacco products. Second messenger signaling pathways will be analyzed with ERK and JNK inhibitors and specific antibodies to ERK and phospho-JNK. Immunoblots using HUVECs will be done to determine expression of ERK/JNK. A better understanding of the detrimental effects that tobacco has on the underlining causes of atherosclerosis can advance the quest of controlling the disease.

Mentors: L. Jack Windsor, Department of Oral Biology; Richard L. Gregory, Department of Oral Biology, Indiana University School of Dentistry, Indianapolis, IN 46202

This study was funded by the Indiana University-Purdue University Indianapolis Multidisciplinary Undergraduate Research Institute (MURI).

---