TITLE: Fluoride Alters Gene Expression via H3K27Ac Modification in LS8 Cells

PREFERRED PRESENTATION TYPE: Poster

SCIENTIFIC GROUP/NETWORK CATEGORY: Mineralized Tissue

PRESENTER: Melanie Mendonca

PRESENTER (INSTITUTION ONLY): Nova Southeastern University

AUTHORS (FIRST NAME INITIAL LAST NAME): M. Mendonca[1], S. Yamashita[2], M. Okamoto[2], N. Fujiwara[3], E. T. Everett[4], J. D. Bartlett[5], M. Suzuki[2]

AUTHORS/INSTITUTIONS: M. Mendonca, Nova Southeastern University, Davie, Florida, UNITED STATES;

S. Yamashita, M. Okamoto, M. Suzuki, Oral Science and Translational Research, Nova Southeastern University, Fort Lauderdale, Florida, UNITED STATES;

N. Fujiwara, Department of Oral Health Promotion, Tokushima University Graduate School of Biomedical Sciences, Kuramoto, Tokushima, JAPAN;

E.T. Everett, Koury Oral Health Sciences Bldg., Room 5501H, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, UNITED STATES;

J.D. Bartlett, Biosciences, College of Dentistry, The Ohio State University, Columbus, Ohio, UNITED STATES;

ABSTRACT BODY:

Objectives: Previously we reported that fluoride-mediated histone acetyltransferase (HAT) activation contributed to p53 acetylation to promote fluoride toxicity in mouse ameloblast-like cells (LS8). However, the roles of fluoride-mediated HAT activation in histone acetylation and epigenetic regulation of fluoride-mediated gene expressions remain unidentified. The present study demonstrates that fluoride-mediated gene expression is regulated epigenetically via modification of histone acetylation status (H3K27Ac) in transcription start sites (TSS) in LS8 cells.

Methods: LS8 cells were treated with or without 5 mM fluoride for 24 h and subjected to chromatin isolation followed by immunoprecipitation (ChIP) and DNA purification. ChIP assay was performed using an antibody specific for histone acetylation (H3K27Ac) followed by ChIP-sequencing (ChIP-seq) on the Nextseq500 (Illumina). Genes were identified by differential H3K27Ac peaks with fluoride treatment within ± 1 kb from TSS. Identified genes by ChIP-Seq were analyzed by quantitative real-time PCR (q-PCR) to evaluate mRNA expressions.

Results: ChIP-Seq identified that the differential acetylation status of H3K27 in TSS was associated with mRNA expressions (Bax, p21, Mdm2, p53, Bad and Bcl2) that were altered by fluoride. Fluoride increased H3K27Ac peaks in the TSS of Bax, p21 and Mdm2, while H3K27Ac peaks were decreased in the TSS of p53, Bad and Bcl2. qPCR results showed that fluoride treatment increased mRNAs of Bax, p21 and Mdm2.

Meanwhile, fluoride treatment suppressed mRNAs of p53, Bad and Bcl2. The H3K27Ac status (increase or decrease by fluoride) in the TSS of these genes was concordant with mRNA expressions.

Conclusions: In the present study, we demonstrated for the first time, that gene expression altered by fluoride was epigenetically regulated via H3K27Ac in the TSS in LS8 cells. Our results warrant further investigation to

elucidate epigenetic regulation in fluoride toxicity to develop a potential novel strategy targeting histone modification.

KEYWORDS: Fluoride, HAT, H3K27, Acetylation, Epigenetics.

AWARDS APPLICATIONS: AADOCR Award-AADOCR Hatton Competition

PRESENTER'S STUDENT STATUS: College/University/Pre-Dental or Secondary Student

SUPPORT FUNDING/GRANT NUMBER: NIH grant 1t34gm145509-01a1; NIH grant R01DE027648; NIH grant K02DE029531

FINANCIAL INTEREST DISCLOSURE: NONE

Group Author Abstracts: (none)

ONE SENTENCE SUMMARY: Fluoride alters gene expressions via histone acetylation modification