The College of Graduate Studies and the College of Medicine & Health Sciences Cordially Invite You to a

Master Thesis Defense

Entitled

DECIPHERING THE SALIVARY MICROBIOME IN CROHN'S DISEASE PATIENTS WITH DIFFERENT FACTORS CONTRIBUTING TO DYSBIOSIS

by

Hala Elzayat

Faculty Advisor

Dr. Farah Al Marzooq, Department of Microbiology & Immunology

College of Medicine & Health Sciences

Date & Venue

12 pm

Wednesday, 18 January 2023

YANAH Theatre (2C010), CMHS

Join with ZOOM

Abstract

Crohn's disease (CD) is a chronic inflammatory bowel disease (IBD), common in the UAE. Microbiota is necessary to maintain a balanced gut environment which is essential for good health. Dysbiosis can predispose to many diseases including CD. The oral cavity has the second largest and most diverse microbiota after the gut harboring over 700 species of bacteria. This study aims to investigate the alterations in the salivary microbiome in patients with CD compared to healthy controls (HC). It also aims to compare CD patients for salivary microbiome complexity and diversity according to different factors that can contribute to dysbiosis, including oral health, IBD drug use, disease duration, activity of the disease and relapse of symptoms. Finally, it aims to find any correlation between the inflammatory biomarkers in CD with their levels in saliva, and any possible link to oral dysbiosis.

A total of 80 saliva samples were collected from CD patients and HC (n=40 in each group) seeking healthcare from two hospitals in Abu Dhabi, UAE. Information related to the participants' oral and general health was recorded. DNA was extracted from saliva and sequenced using Oxford nanopore technology for salivary microbiome profiling. Salivary supernatant was used to measure inflammatory biomarkers including C-reactive protein (CRP) and calprotectin (CAL) by enzyme-linked immunosorbent assay (ELISA). Data was analyzed using appropriate bioinformatics and biostatistics tools.

Obvious differences in the salivary microbiome of CD were found when compared to HC. Five dominant species were enriched in CD and depleted in HC, namely *Veillonella dispar*, *Megasphaera stantonii*, *Provetella jejuni*, *Dolosigranulum pigrum* and *Lactobacillus backii*. Oral health is confirmed to have paramount significance in the dysbiosis of the oral microbiota since most significant features are cariogenic such as *Streptococcus mutans* or periopathogenic such as *Fusobacterium periodonticum*. Loss of operational taxa diversity was shown by multiple alpha diversity indices, as well as dissimilarities between CD samples that were interpreted through beta diversity measures. The activity of the disease, duration and the relapse of symptoms also had great impacts on the shift or disruption of the normal balance of the oral microbiota. Interestingly, treatment with biologicals led to the emergence of a novel species called *Simonsiella muelleri*. When immunomodulatory agents were used in conjunction with biologicals, pathogenic species such as *Salmonella enterica, Escherichia coli, Klebsiella pneumoniae* and *Pseudomonas aeruginosa* were recognized. Finally, inflammatory biomarkers were also analyzed confirming an association with significance of being biomarkers for the presence of inflammatory bowel disease and reduction of diversity in the oral microbiome.

In conclusion, we were able to decipher the salivary microbiome of CD patients and prove that the interplay of variable factors contributed to dysbiosis. Each factor seems to have a unique effect on the oral microbiome. Nevertheless, oral health status was found to be of greatest impact. Poor oral health contributes to oral dysbiosis and hence can induce bowel inflammation, especially in the presence of oral periodontal disease such as periodontitis which is obviously an inflammatory condition. Oral health had the greatest impact according to the hypothesis of the ingestion of the tremendous amount of saliva being a reservoir of different microbial species (pathogenic or opportunistic), contributing to dysbiosis in CD patients. In addition, IBD drugs had equivalent influence as the oral health in terms of dysbiosis. Saliva can be used as a tool to detect bacterial dysbiosis and some degree of inflammation, since it is less invasive and more convenient.

Our study is considered unique as this type of in-depth salivary microbiome analyses in CD is established for the first time in the UAE, utilizing a sequencing technique with high resolution enabling the characterization of microbiota down to the species level, in addition to the involvement of multiple factors that added to its uniqueness.

Keywords: Crohn's disease, inflammatory bowel disease, microbiota, microbiome, dysbiosis, IBD drugs, diversity, inflammatory biomarkers, C-reactive protein (CRP) and calprotectin (CAL).