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DoxyPrEP Impact on the Microbiome of Men Who Have Sex With Men and Transgender Women on HIV PrEP

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BACKGROUND

- Doxycycline post-exposure prophylaxis (doxyPEP; *200mg within 72h of exposure*) now has established efficacy for preventing bacterial sexually transmitted infections (STI) in gay, bi, and other men who have sex with men (GBM) and transgender women (TGW)[1-3].
- Doxycycline pre-exposure prophylaxis (doxyPrEP; *100mg daily*) has shown potential in pilot studies in reducing STI in these same populations[4-6].
- A recent study showed no impact from doxyPEP on the compositional microbiome[7]; however, doxyPrEP impact is unknown.
- This study assessed rectal microbiome changes over 48 weeks in participants on HIV PrEP enrolled in a doxyPrEP trial.

METHODS

- The Dual Daily HIV and STI PrEP (DuDHS) study is an open-label, randomized pilot trial comparing immediate vs. deferred doxyPrEP (100mg daily) in GBM and TGW on HIV PrEP with prior syphilis over 48 weeks. The immediate arm received 48 weeks of doxyPrEP; the deferred arm received 24 weeks, starting at week 24.
- Participants (n=52) were recruited from a clinic in Vancouver, Canada
- Microbial DNA data was generated by 16S rRNA sequencing from 123 rectal swabs available from 41 participants that collected baseline (BL), week 24, and week 48 samples. Microbiome changes were evaluated using Wilcoxon signed rank and Mann-Whitney U tests, with p-values adjusted for multiple comparisons (FDR=5%).

RESULTS

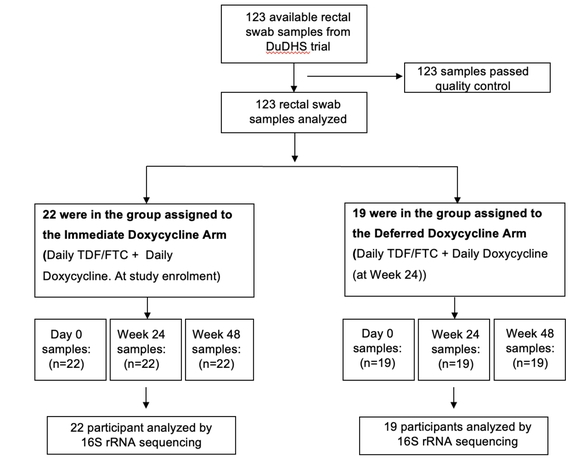


Figure 1: Study design and 16S rRNA sequencing analysis of DuDHS participants. TDF/FTC: tenofovir disoproxil fumarate/emtricitabine

Over 48 weeks, doxyPrEP use in men who have sex with men and transgender women had minimal impact on the compositional microbiome. Further research is needed to explore the impact of doxyPrEP/PEP on microbiome function and antimicrobial resistance.

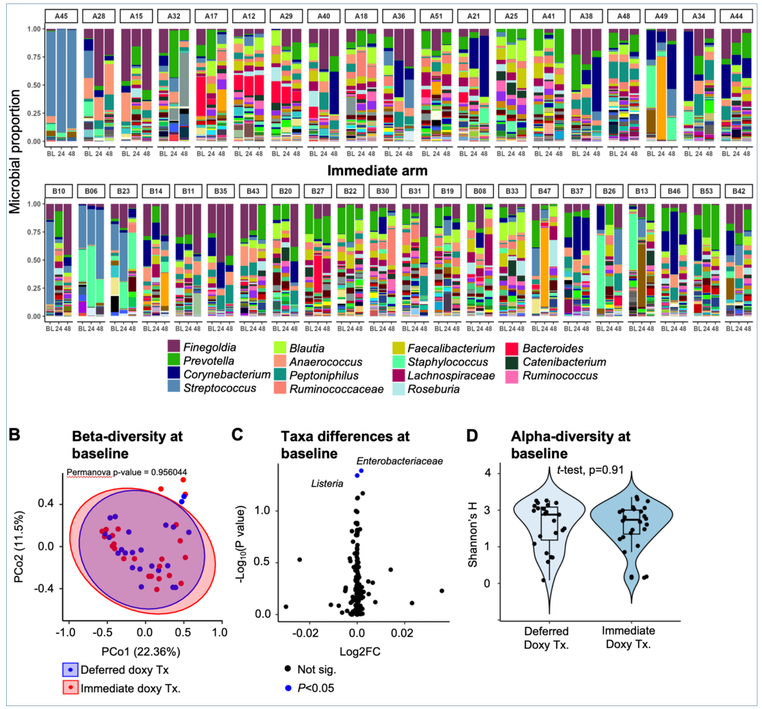


Figure 2. Rectal microbiome composition of study participants in the DuDHS study. The composition of the rectal microbiome was determined using 16S rRNA sequencing. (A) Microbial composition of rectal samples of each study participant in each study arm (Immediate (n=22) and Deferred (n=19)) at their baseline visit (BL), week 24, and week 48. Different colors represent the predominant microbial taxa. (B) Beta-diversity of baseline microbiome profiles of study participants in each study arm (Bray-Curtis distance metric). Two-sided PERMANOVA p value is shown. (C) Differential abundance analysis of individual taxa in each study arm at baseline. Blue dots represent those $P<0.05$. (D) Alpha diversity (Shannon's H) of each study arm at baseline visit. Differences between groups in C and D were determined by Mann-Whitney *U* test adjusting for multiple comparisons. Taxa abundance of each sample is the average of two technical replicates.

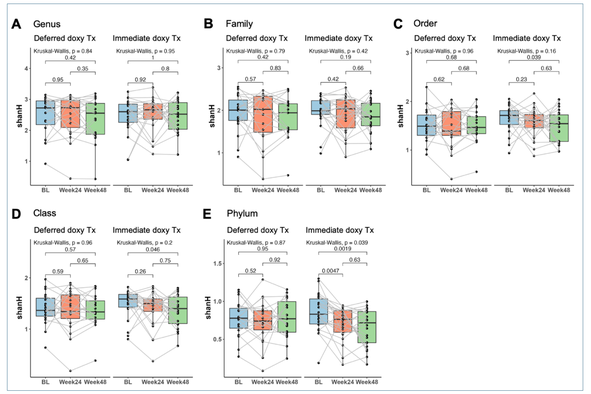


Figure 3. Effect of doxycycline on the rectal microbiome alpha diversity in DuDHS study participants over 24 and 48 weeks. Rectal microbiome composition was evaluated from rectal swab samples collected at baseline, 24 and 48 weeks after randomization to either the immediate (n=22) or deferred (n=19) doxyPrEP study arms. A comparison of alpha diversity measures (Shannon's H) over time in each study arm are shown at the genus (A), family (B), order (C), class (D), and phylum level (E). Box plots show the median, upper and lower quartiles. Differences between follow-up timepoints were determined Wilcoxon signed rank tests in paired comparisons, and across all 3 timepoints using Kruskal-Wallis test.

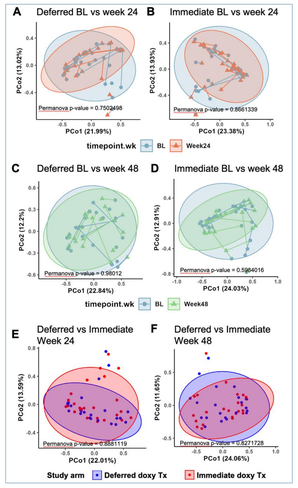


Figure 4. Effect of doxyPrEP on the rectal microbiome beta diversity in DuDHS study participants over 24 and 48 weeks. Rectal microbiome composition was evaluated using 16S rRNA sequencing of rectal swab samples collected at baseline, 24 and 48 weeks after randomization to either the immediate or deferred doxycycline study arms. A comparison of beta diversity measures (Bray-Curtis) over time within each study arm (A-D) and between study arms (E-F) are shown. The distance between triangles and circles reflects the relative change in microbiome composition, and degrees of significance represented by two-sided PERMANOVA p values. (A) Deferred doxyPrEP arm, baseline vs. week 24; (B) Immediate doxyPrEP arm, baseline vs week 24; (C) Deferred doxyPrEP arm, baseline vs. week 48; (D) Immediate doxyPrEP arm, baseline vs week 48. BL = baseline; (E) Deferred vs. Immediate arm, week 24; (F) Deferred vs Immediate arm, week 48.

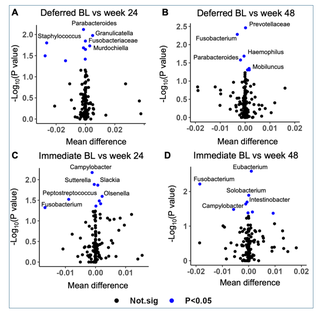


Figure 5. Changes in abundance of individual microbial taxa over time in rectal mucosa of study participants. The composition of the rectal microbiome was determined using 16S rRNA sequencing. Differential abundance analysis of microbial taxa within each participant was performed between timepoints in each study arm (Deferred and Immediate arms) using Wilcoxon signed rank tests. Volcano plots show relative abundance of individual microbial taxa differences (genus) along the x-axis, with the p-value statistic along the y-axis. Dots in blue are those that were below a p value significance threshold of 0.05, although none passed a 5% FDR.

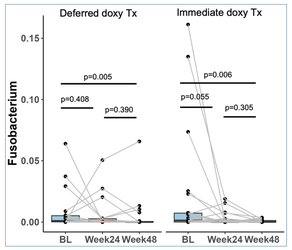


Figure 6. Changes in abundance of *Fusobacterium* in the rectal mucosa of study participants in the DuDHS study. This plot shows the change in relative abundance of *Fusobacterium* between timepoints in each participant within each study arm (Deferred and Immediate arms) using Wilcoxon signed rank tests. BL = baseline.

CONCLUSIONS

- To our knowledge, this is the first study to prospectively examine compositional microbiome changes in GBM and TGW using doxyPrEP.
- Our results show that daily doxycycline use had minimal impact on the rectal microbiome over 48 weeks.
- Given the increasing use of doxyPEP and doxyPrEP for STI prevention, and the concerns around antimicrobial resistance impacts and human microbiome disruptions, future work must prioritize longitudinal impacts of doxyPEP/PrEP on microbiome function, with particular emphasis on the resistome (i.e. antibiotic resistance genes).

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