Colonic microbiota exhibits disparate associations with HIV infection and sexual practices

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Reported by Jules Levin

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Conclusions

- Treated HIV infection exerts a robust effect on the gut bacterial community across human subject populations, in both men and women, that pervades sexual practice
- MSM microbiota signature is unique and does not overlap with that of HIV infection
- MSM represents a strong confounding variable in PLWH study population
- Receptive anal intercourse, regardless of condom use, is linked with specific gut bacterial communities, in both men and women, which may represent a driving factor for the consistently observed MSM-associated *Prevotella*-rich community
- HIV-associated microbiota signature correlates with CD4 nadir and prevalent non-communicable comorbidities

Objective: Effective antiretroviral therapy (ART) has prolonged survival and shifted the morbidity spectrum for people living with HIV (PLWH) from AIDS-associated opportunistic infections and malignancies towards age-associated non-communicable comorbidities (AANCCs), with these being more prevalent in PLWH

compared with in age-matched HIV-uninfected individuals. A key contributor to the current disease spectrum includes HIV-associated inflammation and immune activation, the aetiology of which in PLWH remains incompletely defined. Gut microbial dysbiosis is thought to be a potential important contributor, but data thus far are conflicting regarding the role that lifestyle factors, including sexual orientation and behaviour, and HIV-infection itself have on gut microbial dysbiosis.

Methods: Using 16S rRNA gene sequencing, we profiled the microbiota from fecal samples of PLWH with suppressed viraemia on ART and HIV-uninfected controls participating in the AGEhIV Cohort Study. PLWH were selected to include 40 men having sex with men (MSM),
20 men having sex with women (MSW) and 20 females (F) matched 1:1 by age, sex, sexual orientation, BMI, birth country and smoking status with HIV-uninfected controls.

Results: HIV-infection was associated with alterations in the gut microbiota including an enrichment in *Enterobacteriaceae* and *Desulfovibrionaceae* members and a depletion of short chain fatty acids-producing bacteria such as *Lachnospiraceae* and *Ruminococcaceae*. Furthermore,
comparisons between MSM and non-MSM males revealed a **unique MSM-associated micro biome signature** characterized by an enrichment particularly in *Prevotellaceae* members (Figure 1), which was independent of HIV-infection. Finally, **practicing receptive anal intercourse, regardless of condom use, was linked to a specific bacterial community variance independently of sex, which may explain the *Prevotella*-rich microbiome in MSM**.

Conclusions: Our data provide unique evidence that colonic microbiota exhibit disparate associations with HIV-infection and sexual practices.

Colonic microbiota exhibits disparate associations with HIV-infection and sexual practices

Eveline Verheij

Abstract number: ADRLH-42

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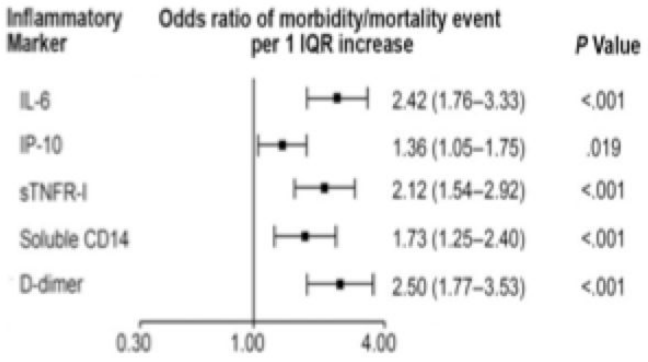
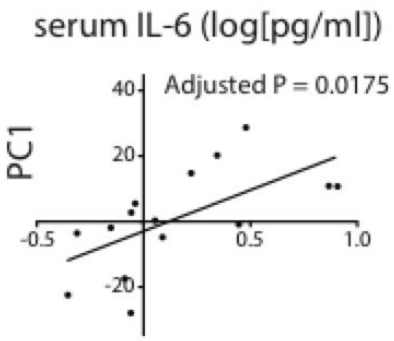
Comorbidities in treated HIV infection are linked to inflammation



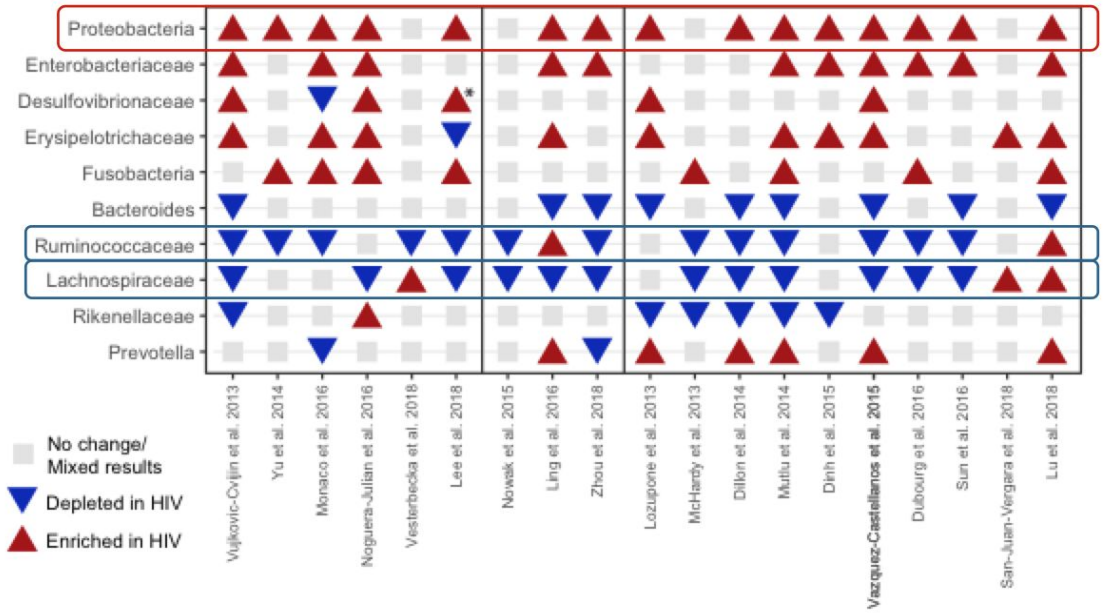
Adapted from Tenorio et al. JID 2014



Vujkovic-Cvijin et al. STM 2013

Studies show that comorbidities are associated with inflammation, and that inflammation is associated with the gut micro biome.

Microbiota shifts in HIV infection



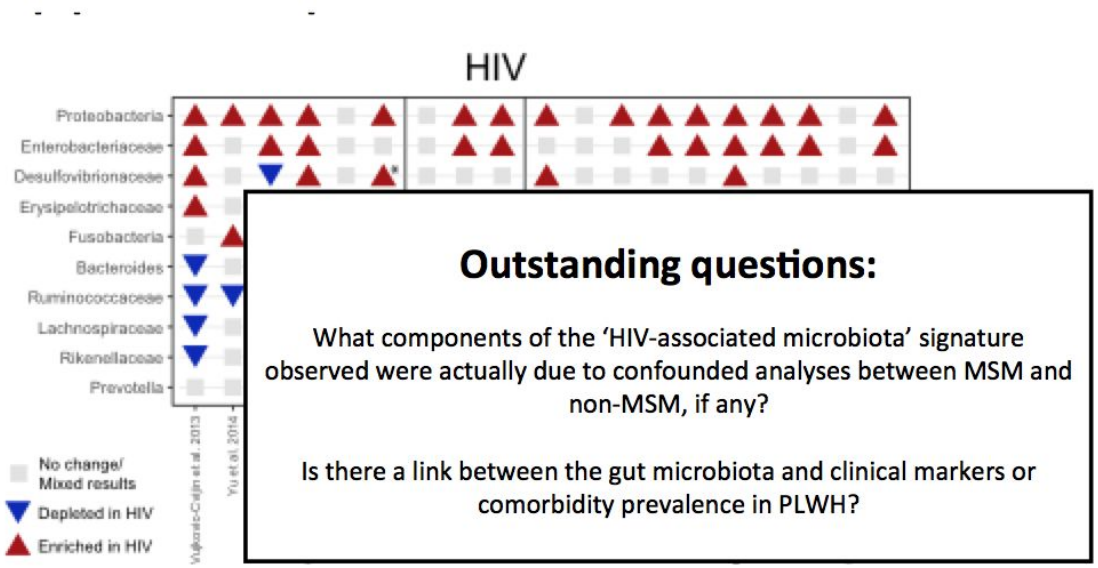
Vujkovic-Cvijin & Somsouk,
Current HIV/AIDS Rep 2019

But what do we know about the microbiome of people living with HIV.
In this figure, every column represents a different study, and each row shows which specie is either enriched - indicated by the red arrows - or depleted -as indicated by the blue arrows - in people living with HIV.

Pathogenesis

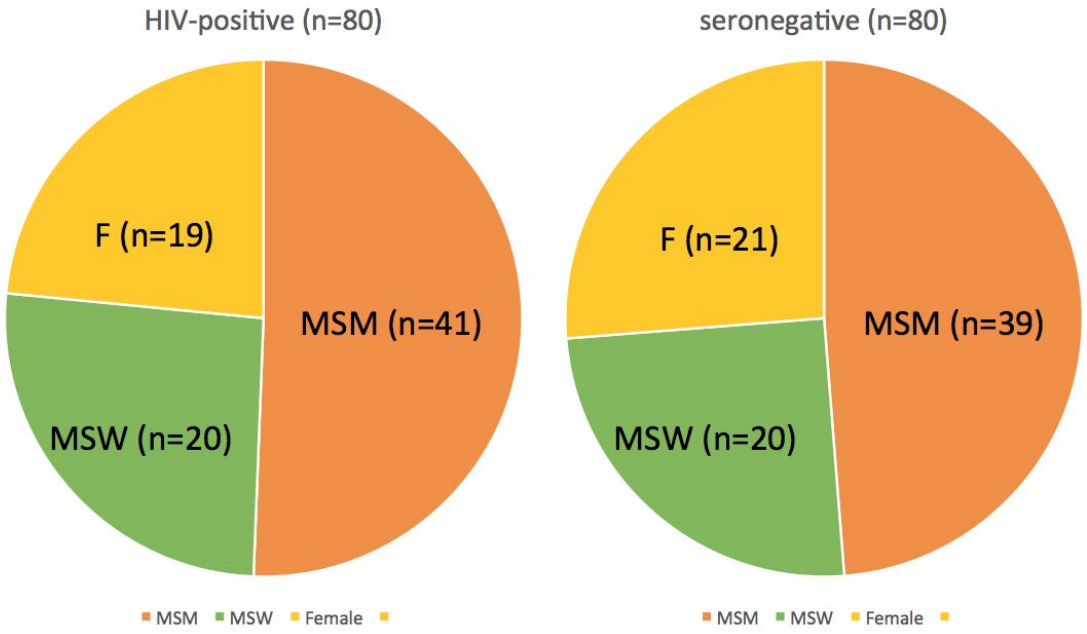
What are the mechanisms behind the association between the microbiome and HIV-associated inflammation?
First of all, translocation of Proteobacteria is known to activate systemic immunity.
Depletion of Clostridiales is associated with less producion of short chain fatty acids that maintain a healthy gut epithelium. Its also associated with increased disease serverity in Crohn's disease and leads to experimental colonic inflammation.

Microbiota shifts in HIV infection



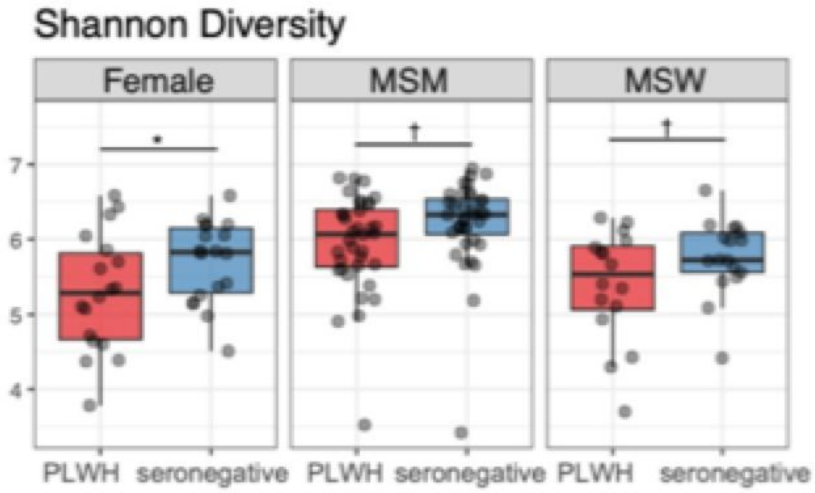
However, microbiota shifts in HIV infection, appears to parallel shifts in MSM versus nonMSM, possibly driven by the fact that the HIV positive populations consists mostly of MSM and the majority of the seronegative population is non-MSM.
Begs the question are the differences we've found due to HIV infection or due to MSM status?
And the second question I would like to answer is whether the gut microbiota is linked to clinical markers and comorbidity prevalence in PLWH.

No differences in age, BMI, country, and smoking status



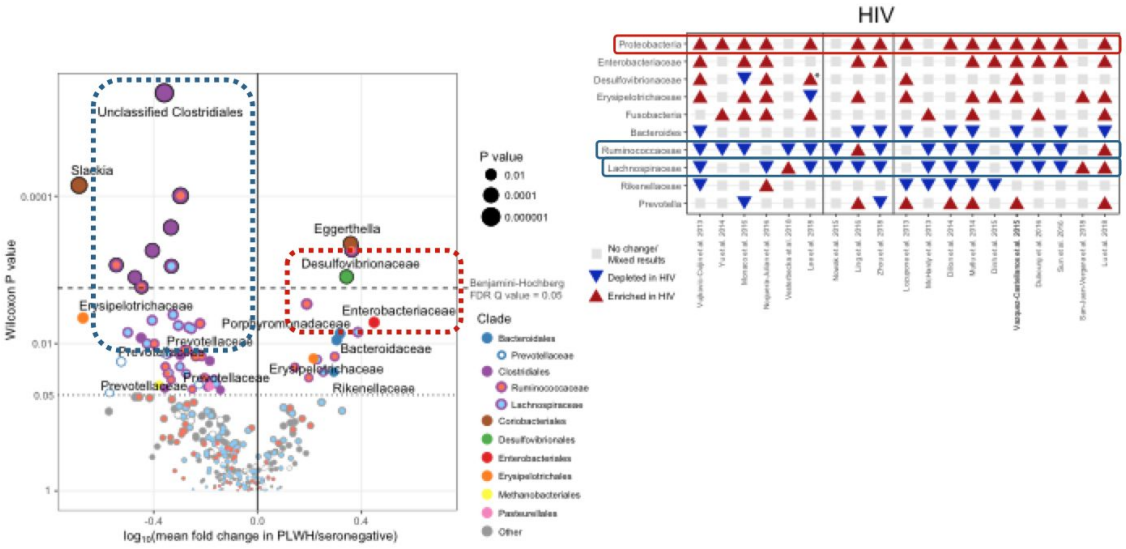
In order to answer those questions, we actively selected from our AGEHIV study cohort PLWH who are chronically infected, with suppressed viremia on antiretroviral therapy and matched them 1 on 1 with HIV-uninfected controls.
We created 3 subgroups, MSM, men who have sex with women (MSW) and females. They were all matched 1:1 on age, body mass index, and country of birth as we not from other studies that these variable have an impact on the microbiota.

Alpha diversity is ***decreased*** in PLWH

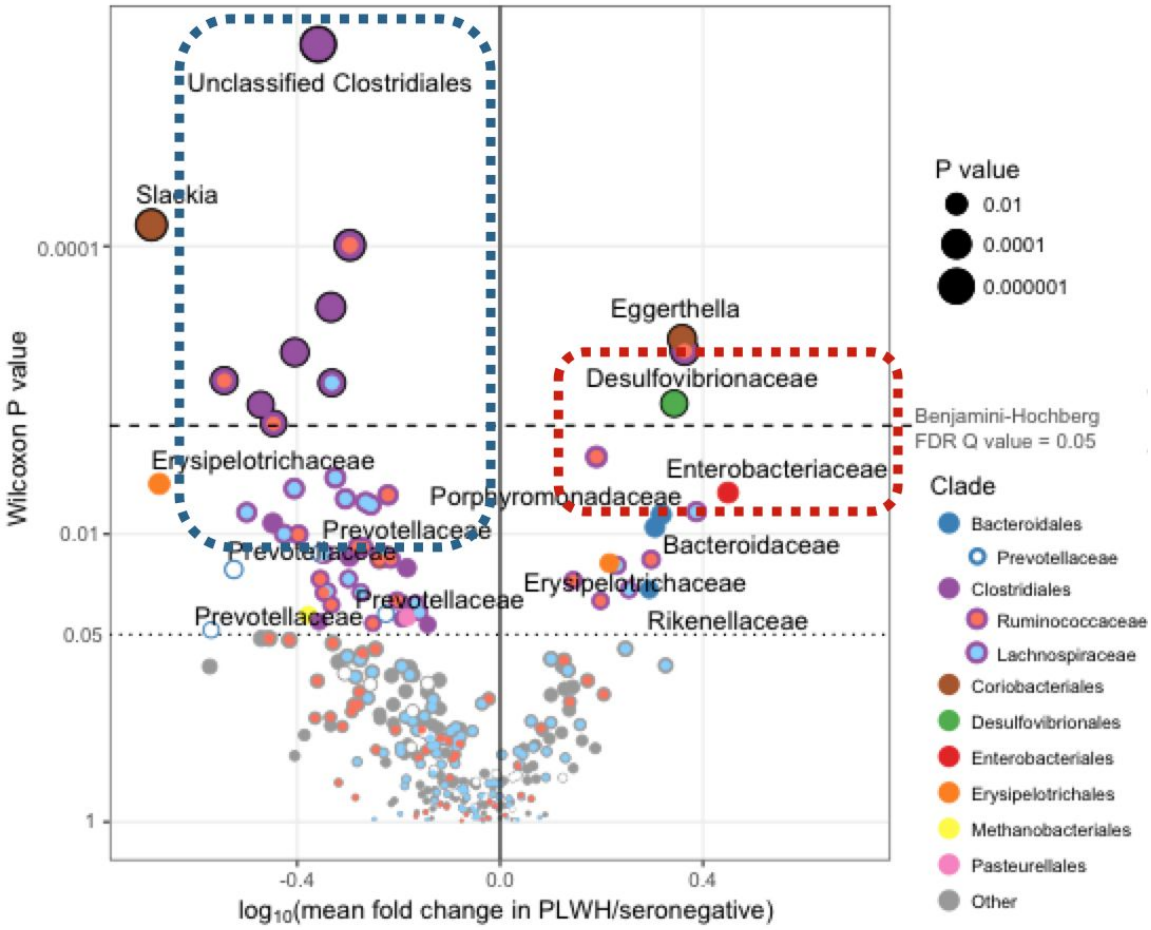


Turning to the results: for each different group I'm going to discuss their microbiome.
First we looked at the alpha diversity. Alpha diversity is a measure of how many different bugs are present in the gut.
We see that in PLWH, the alpha diversity is decreased in compared to seronegative controls in all three subgroups.
Secondly, we looked at beta diversity.
The beta diversity is a measure of how different the microbial gut composition is, as compared to another.
It reveals clustering of PLWH and HIV-negative controls in all three subgroups.

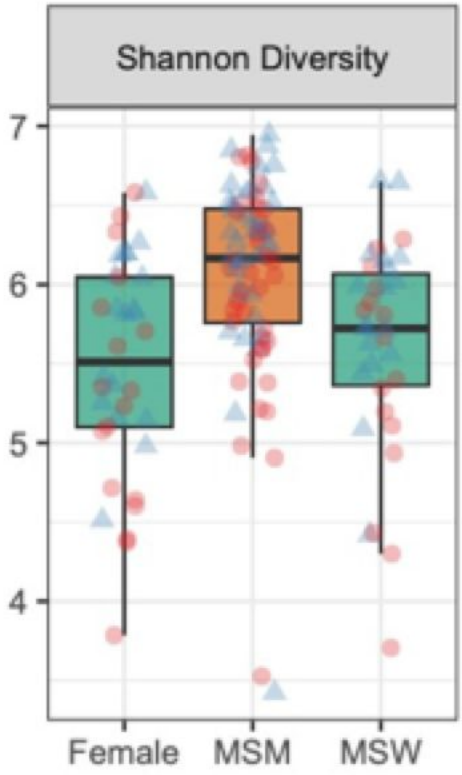
The microbiota signature in PLWH mirrors that of prior studies



Importantly, our results mirror that of previous studies. Looking at this vulcanoplot, which shows on the right side of the figure the taxa enriched in PLWH and on the left site the taxa depleted in PLWH as compared to seronegative controles. You can see that Proteobacteria (in red) are enriched and Clostridiales (in blue) are depleted.

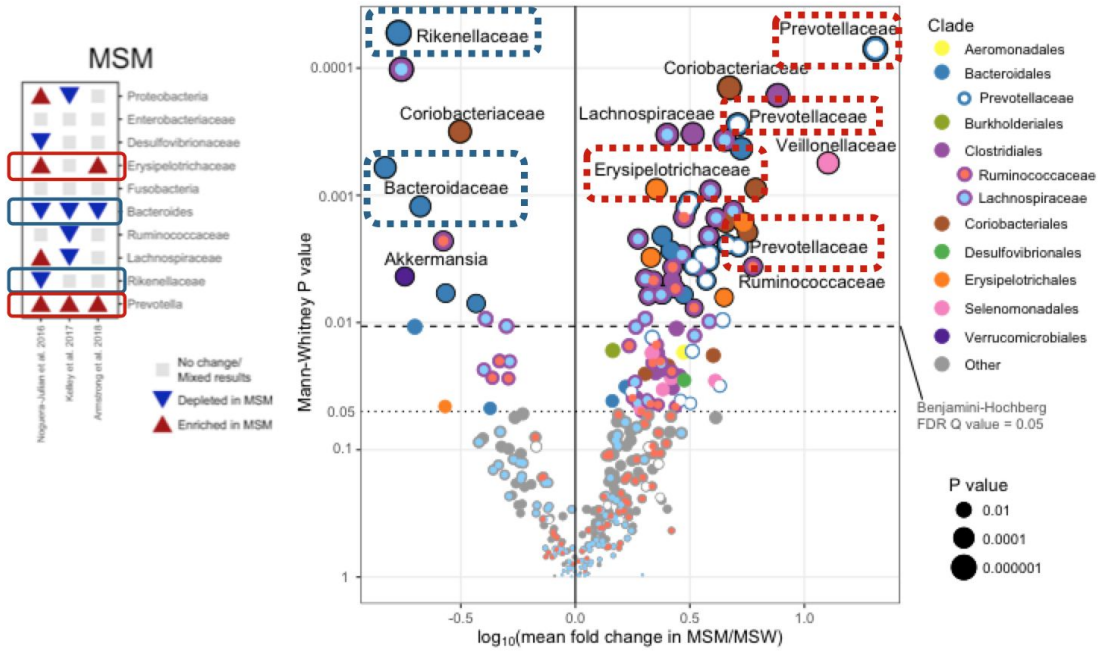

Unclassified Clostridiales
Slackia
Eggerthella
Desulfovibrionaceae
Erysipelotrichaceae
Enterobacteriaceae
Porphyromonadaceae
Prevotellaceae
Prevotellaceae
Bacteroidaceae
Erysipelotrichaceae
Prevotellaceae
Prevotellaceae
Rikenellaceae
Wilcoxon P value
0.0001
0.01
0.05
1
-0.4
0.0
0.4
log10(mean fold change in PLWH/seronegative)
Benjamini-Hochberg
FDR Q value = 0.05
P value
0.01
0.0001
0.000001
Clade
Bacteroidales
Prevotellaceae
Clostridiales
Ruminococcaceae
Lachnospiraceae
Coriobacteriales
Desulfovibrionales
Enterobacteriales
Erysipelotrichales
Methanobacteriales
Pasteurellales
Other

Alpha diversity is ***increased*** in MSM



What do we see if we look at MSM?
Looking at MSM versus MSW and females, showed in increased alpha diversity in MSM. And the microbiome composition is different from MSM versus MSW

MSM vs MSW microbiota signature is distinct

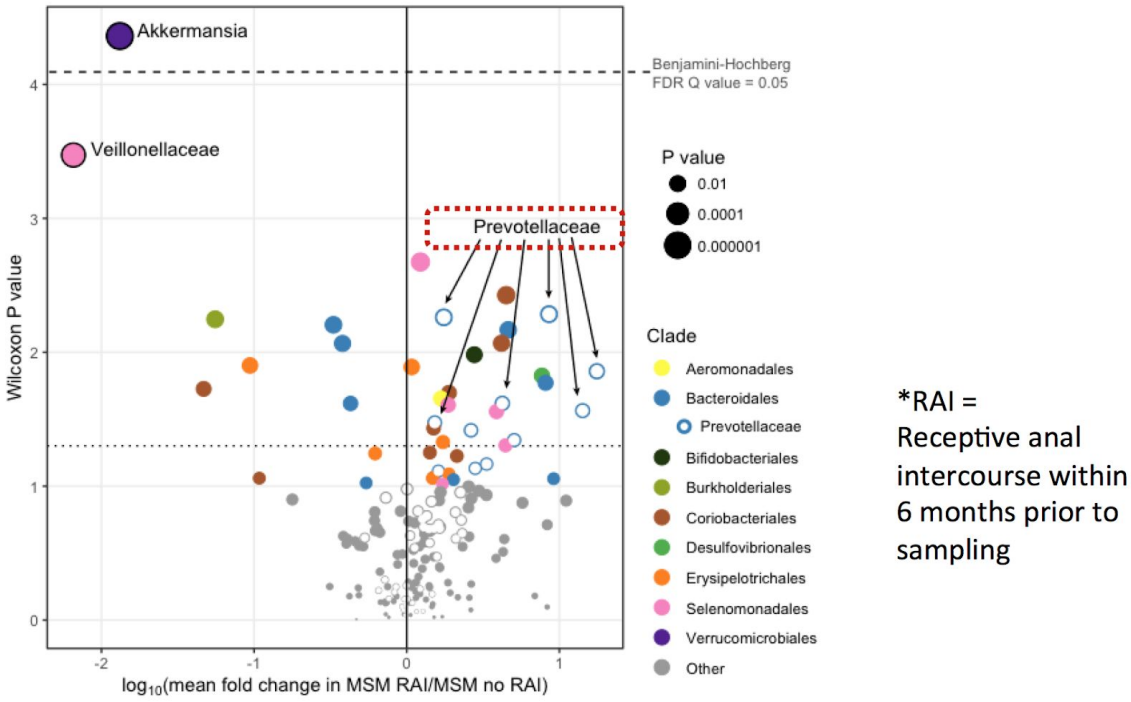


Looking at this volcano-plot, on the right the species enriched in MSM and on the left species depleted in MSM shows that our results again mirror those of prior studies. Showing enrichment of Prev in MSM and depletion of Bact.

We were of course interested whether the same taxa observed to be enriched in PLWH are the same as those enriched in MSM or conversely if those depleted in PLWH were also depleted in MSM.

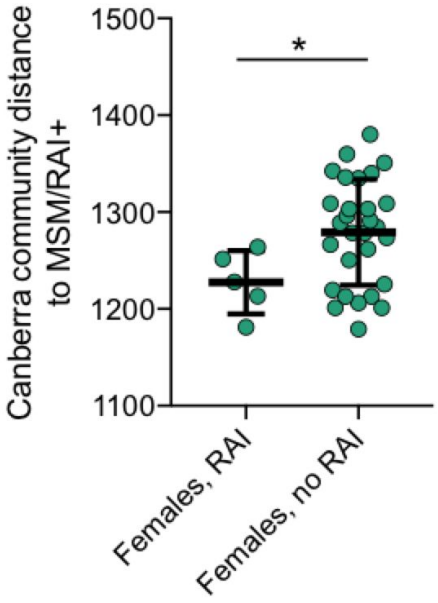
In order to do this, we selected the taxa that differed significantly between PLWH and seronegative controls and MSM versus MSW. When we plotted the taxa that overlapped, we surprisingly, saw that these taxa had opposing abundance trends. As you can see from the figure on the right you can see the taxa enriched in MSM and PLWH and the did not overlap. In fact, those enriched in MSM (indicated by the purple arrow) were depleted in HIV (mint green arrow). And vice versa.

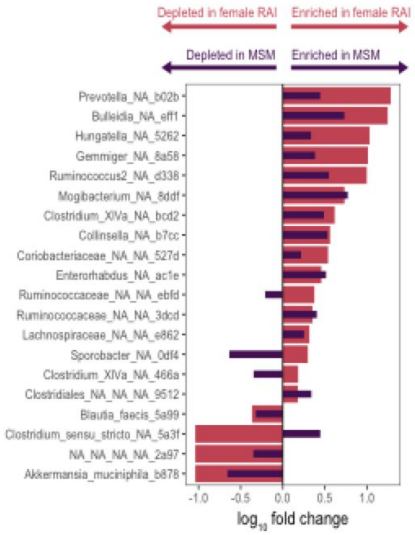
Microbiota shifts in MSM with recent receptive anal intercourse (RAI) vs. MSM without RAI resemble shifts of MSM vs MSW



We tried to look if having recent anal intercourse had anything to do with the signature. Using the same trick as before, we looked at MSM who reported recent intercourse and we compared them with those without intercourse, and you can see that the shifts resemble that of MSM versus non-MSM.

Microbiota of females engaging in RAI bears resemblance to that of MSM



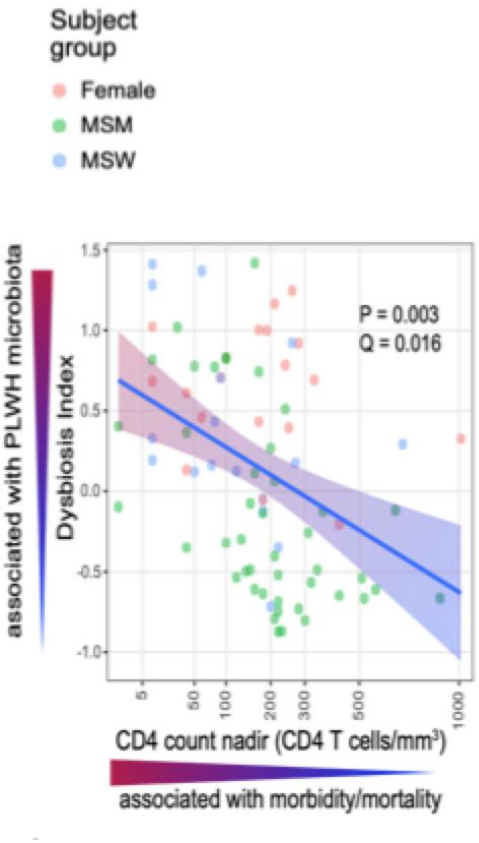


- Bacteria enriched in Females that engaged in RAI overlap with bacteria enriched in MSM

Shown are taxa with P<0.05 (Mann-Whitney test) difference in abundance in both of the following comparisons: F/RAI+ vs F/RAI-, MSM/RAI+ vs MSM/RAI-

Females practicing RAI, illustated the same trend as MSM who report RAI .
The figure on the lefts shows that the difference of the microbiome of females RAI is more similar to that of MSM/RAI +, females without RAI. Moreover, the taxa enriched in Females with RAI overlapped with taxa enriched in MSM with RAI and again vice versa.

HIV-associated microbiota signature correlates with CD4 nadir



To understand the clinical impact of the HIV-associated microbiome, we constucted a measure which is called the Dysbiosis Index, in which we collapsed into a single number, the shifts in bacterial taxa that were characteristic of PLWH versus seronegative controls. A higher number indicated higher dysbiosis.

So, DI is associated with lower CD4 nadir. Additionaly, the index is associated with higher number of prevalent comorbidities, in all three HIV-infected subgroups.
And this effect is independent from CD4 nadir.

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