Effect of High-Risk Sexual Behaviour on Diversity of the Vaginal Microbiota and Abundance of *Lactobacillus*


3rd International Workshop on Microbiome in HIV Pathogenesis, Prevention, and Treatment

Friday 20th October, 2017
The Vaginal Microbiome

- Vaginal epithelium lined with bacteria

- “Healthy” vaginal microbiome thought to be dominated by Lactobacillus species and low pH (~4.0)

- “Unhealthy” vaginal microbiome is polymicrobial

- Dysbiosis can lead to bacterial vaginosis

Modified from: Burgener et al., 2015
The Species Present in the Vaginal Microbiota Depend on Ethnicity

African American Women

Women of European Ancestry

Modified from: Fettweis et al., 2014
The Vaginal Microbiome

- A greater variety of bacterial species in the Vaginal Microbiome is associated with negative reproductive outcomes (HIV, STIs, PID, preterm birth)

- Bacterial vaginosis is consistently linked with increased risk of STIs

Modified from: Borgdorff et al., 2014
The Vaginal Microbiome

Lactobacillus-Deficient Cervicovaginal Bacterial Communities Are Associated with Increased HIV Acquisition in Young South African Women

Christina Gesmann,1,2 Melis N. Anahtar,1,2 Scott A. Handley,3 Mara Farcașanu,1 Caleb Abu-Ali,4 Brittany A. Bowman,1 Nikita Padavattan,6 Chandni Desai,9 Lindsay Droit,2 Amber Moodley,6 Mary Dong,1,4 Yuezhou Chen,7 Nasreen Ismail,5 Thubzi Ndung’u,1 Musia S. Ghebre- Michael,1,2 Duane R. Wesemann,7 Caroline Mitchell,8 Krista L. Dong,1,10 Curtis Huttenhower,3 Bruce D. Walker,1,5,9,11,12 Herbert W. Virgin,3 and Douglas S. Kwon1,9,10,12,*

Cervicovaginal Bacteria Are a Major Modulator of Host Inflammatory Responses in the Female Genital Tract

Mëlis N. Anahtar,1,2 Elizabeth H. Byrne,1 Kathleen E. Doherty,1 Brittany A. Bowman,1 Hidemi S. Yamamoto,9 Magali Sourniaim,1 Nikita Padavattan,6 Nasreen Ismail,5 Amber Moodley,6 Mary E. Sobatni,2 Musia S. Ghebre- Michael,1 Chad Nixbaum, Curtis Huttenhower,3 Herbert W. Virgin,3 Thubzi Ndung’u,1,4,10 Krista L. Dong,1,10 Bruce D. Walker,1,5,12 Raina R. Fichorova,3 and Douglas S. Kwon1,9,12,*

Relationship between vaginal microbial dysbiosis, inflammation, and pregnancy outcomes in cervical cerclage

Lindsay M. Kindlinger,1,2,3 David A. MacIntyre,4,5 Yun S. Lee,1 Julian R. Marchesi,6,5 Ann Smith,5 Julie A. K. McDonald,4 Vasso Terzidou,6,4 Joanna R. Cook,1 Christoph Lees,1,2,7 Fidan Israf- Bayli,8 Yazmin Faiza,9 Philip Toots-Hobson,9 Mark Slack,6 Stefano Cacciapuoti,7 Elaine Holmes,4,10 Jeremy K. Nicholson,9,10 T. G. Teoh,9 Phillip R. Bennett1,2,*

Lactobacillus crispatus Dominant Vaginal Microbiome Is Associated with Inhibitory Activity of Female Genital Tract Secretions against Escherichia coli

Jeny P. Gharley1,9,*, Benjamin C. Smith2,9, Ziguí Chen2, Niall Buckley2, Yungtai Lo3, Adam J. Ratner4, Betsy C. Herold1,2,3,9, Robert D. Burk1,2,3,9

Cervical intraepithelial neoplasia disease progression is associated with increased vaginal microbiome diversity

A. Mitra1, D. A. MacIntyre1, Y. S. Lee1, A. Smith1, J. R. Marchesi1, B. Lehne1, R. Bhatia6, D. Lynn2, E. Paraskevaidis1, I. V. L2, E. Holmes1, J. K. Nicholson2, P. R. Bennett1,2 & M. Kyrgiou1

Bacteria in the Vaginal Microbiome Alter the Innate Immune Response and Barrier Properties of the Human Vaginal Epithelia in a Species-Specific Manner

Sylvie Y. Deedrlinger1, Andrea L. Throp2, and Melissa M. Herbst-Kralovetz2
The Vaginal Microbiome

From: Burgener et al., 2015
Research Question

- Does sex work alter the vaginal microbiome?

Aim

- Profile the vaginal microbiome of women engaged in high-risk sexual behaviour (Sex Workers) with women who are not engaged in high-risk sexual behaviour (Non-Sex Workers) from the same community.

Hypothesis

- Sex Workers will have greater bacterial diversity in their vaginal microbiome as compared to Non-Sex Workers.
- Further, these two groups of women will have distinct bacterial profiles by 16S sequencing.
Study Framework and Design

- Retrospective Clinical Study
- In Collaboration with Universities of Manitoba and Nairobi
- Nairobi, Kenya (Pumwani Clinics)
  - Sex Worker Cohort
  - Non-Sex Workers
- Banked cervicovaginal lavage
- Sex Workers N=48
  - New Sex Workers
    (<3 years of sex work)
- Non-Sex Workers N=19
## Participant Characteristics

<table>
<thead>
<tr>
<th></th>
<th>Non-Sex Workers</th>
<th>Female Sex Workers</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Total</strong></td>
<td>N=19</td>
<td>N=48</td>
<td></td>
</tr>
<tr>
<td><strong>Mean Age (Range), Years</strong></td>
<td>29.8 (19-45)</td>
<td>31.1 (21-45)</td>
<td>0.5</td>
</tr>
<tr>
<td>Unknown</td>
<td>1</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td><strong>Marital Status:</strong></td>
<td></td>
<td></td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Married and Living with a Man</td>
<td>9 (47%)</td>
<td>0 (0%)</td>
<td></td>
</tr>
<tr>
<td>Married but not Living with a Man</td>
<td>3 (16%)</td>
<td>7 (15%)</td>
<td></td>
</tr>
<tr>
<td>Unmarried but Living with a Man</td>
<td>2 (11%)</td>
<td>0 (0%)</td>
<td></td>
</tr>
<tr>
<td>Unmarried</td>
<td>4 (21%)</td>
<td>37 (77%)</td>
<td></td>
</tr>
<tr>
<td>Unknown</td>
<td>0 (0%)</td>
<td>4 (8%)</td>
<td></td>
</tr>
<tr>
<td><strong>Menstrual Cycle Stage:</strong></td>
<td></td>
<td></td>
<td>0.03</td>
</tr>
<tr>
<td>Proliferative</td>
<td>0 (0%)</td>
<td>3 (6%)</td>
<td></td>
</tr>
<tr>
<td>Secretory</td>
<td>1 (5%)</td>
<td>17 (35%)</td>
<td></td>
</tr>
<tr>
<td>Hormonal Contraceptive</td>
<td>8 (42%)</td>
<td>10 (21%)</td>
<td></td>
</tr>
<tr>
<td>Unknown</td>
<td>10 (53%)</td>
<td>18 (38%)</td>
<td></td>
</tr>
<tr>
<td><strong>Nugent Score (%)</strong></td>
<td></td>
<td></td>
<td>0.46</td>
</tr>
<tr>
<td>0-3</td>
<td>10 (53%)</td>
<td>24 (50%)</td>
<td></td>
</tr>
<tr>
<td>4-6</td>
<td>6 (31%)</td>
<td>15 (31%)</td>
<td></td>
</tr>
<tr>
<td>7-10</td>
<td>3 (16%)</td>
<td>9 (19%)</td>
<td></td>
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</tbody>
</table>
Sex Workers have Greater Diversity in their Vaginal Microbiota as compared to Non-Sex Workers from the Same Community

A

B

C

Observed Species (OTUs)

Chao1 Richness Estimator

Shannon Diversity Index
Sex Workers have Significantly Different Vaginal Microbiota than Non-Sex Workers and are Less Likely to have *Lactobacillus* as the Most Abundant Genus.

58% (11/19) vs. 17% (8/48) P=0.002

*L. iners* prevalent in Non-Sex Workers and High Diversity prevalent in Sex Workers.
Vaginal Microbiota Cluster by Relative Abundance of *Lactobacillus*

A

Sex Workers and Non-Sex Workers

B

Relative Abundance *Lactobacillus*
Six Distinct Vaginal Microbiota Clusters were Identified

- Three sub-clusters make up Community State Type (CST) IV (High Diversity)
- BUT... 55% of women with low Nugent Scores (0-3) fall within the High Diversity CST
Diversity of the Vaginal Microbiota may not always be linked to Nugent Score

Concordance between 16S and Nugent Score

- **A** All Women (N=67)
- **B** Non-Sex Workers (N=19)
- **C** Sex Workers (N=48)
- **D** Sex Workers with Nugent Score 0-3 (N=23)

![Graphs showing diversity indices and Nugent scores](image)

**Concordance between 16S and Nugent Score**

- NSW: 82% Concordant, 18% Discordant
- CST IV: 80% Concordant, 20% Discordant
- CST I, II, III: 83% Concordant, 17% Discordant
- FSW: 37% Concordant, 63% Discordant
Conclusions

- Bacterial diversity is greatest in the vaginal microbiome of Sex Workers.
- Women engaged in sex work are less likely to have *Lactobacillus* as the most abundant genus as compared to Non-Sex Workers from the same community.
- Six Vaginal Microbiomes clusters exist within our cohort and they cluster based on the abundance of *Lactobacillus*.
  - 3 Sub-clusters exist within the High Diversity CST IV.
- Nugent Score may not be capable of capturing diversity present in the Sex Worker vaginal microbiota.
- Enhanced diversity and lack of lactobacilli could contribute to increased risk of STIs including HIV in women engaged in high-risk behaviour.

Significance

- ↑ Species Diversity
- ↑ Dysbiosis
- ↑ Inflammation & Barrier Breakdown
- ↑ Susceptibility to HIV
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Lab Staff
Study Participants!!!

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Thank You!

WHY DID THE BACTERIA FAIL ITS MATH TEST?

IT THOUGHT MULTIPLICATION WAS THE SAME AS DIVISION