The vaginal microbiome
in health and disease

Jacques Ravel
Institute for Genome Sciences
Dept. of Microbiology and Immunology
University of Maryland School of Medicine

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Lactobacillus spp. are characteristic of vaginal microbiota in “normal” healthy reproductive age women.

Growth of non-indigenous organisms, including pathogens, is restricted.

The mechanisms are unknown: low pH (≤4.5) lactate other organic acids hydrogen peroxide (?) bacteriocins others?

Albert Siegmund Gustav Döderlein, [German obstetrician and gynecologist, 1860–1941], first described the “Döderlein bacillus” in 1894.
The vaginal microbiota through the lifespan

Birth
Sterile then rapidly colonized by *Lactobacillus*

↓

Pre-puberty
Loss of *Lactobacillus* - strict anaerobes

↓

Puberty
Increased estrogen; community shift

↓

Reproductive age
Glycogen metabolized to produce lactic acid

↓

Menopause
Decreased estrogen, less glycogen, less lactic acid

↓

Pregnancy
High estrogen
The vaginal microbiome in asymptomatic healthy women
The vaginal microbiota in asymptomatic women

- Cross sectional study of 410 asymptomatic healthy women*
  - Four ethnic groups equally represented:
    - Caucasian, Black, Hispanic, Asian
  - Self-collected mid-vaginal swabs **
    - Vaginal pH measurements (Inverness VpH glove)
    - Nugent’s Gram stain scores - estimate of vaginal community disturbance
    - Questionnaire (health and sexual history, hygiene...)
  - Determine bacterial community composition by 454 pyrosequencing of barcoded V1-V2 16S rRNA gene

Vaginal community composition

- Five community state types that differ in their microbial composition and abundance.
- Community state type IV lack significant number of *Lactobacillus* - higher diversity.
- Represent 95% of healthy women in these ethnic groups.

Communities of states types dominated by Lactobacillus spp. are found on the four outer vertices (45%).

Communities of state type IV are found at an inner vertex within the tetrahedron (30%).

Intermediate forms are found along the edges of the tetrahedron (25%).

Defines the vaginal community space.

A plot of principle component analysis (PCA) shows distribution of community states in 3-D space.

Graphics generated with inVUE (inVUE.sourceforge.net)
In these healthy, asymptomatic women, CST IV is associated with high Nugent scores and high pH.

How long does this state persist over time? How frequently does the vaginal microbiota of a women is in this state?
The dynamics of the vaginal microbiota

A longitudinal study
Longitudinal studies of the vaginal microbiota

Study design

- 160 women cohort - prospective longitudinal study
  - Daily Self-collected mid-vaginal swabs for 10 weeks
    - Vaginal pH measurements (Inverness VpH glove)
    - Nugent’s Gram stain scores - estimate of vaginal community disturbance
  - Daily diaries
  - Clinical assessment at enrollment, week 5 and 10
  - Determine bacterial community composition by 454 pyrosequencing of barcoded V1-V3 16S rRNA gene of 50 women
Longitudinal profiles - stability

Phylogenetic abundances (%) of different bacterial types over time.

100 - 0

Weeks

0 - 10

Phylogenetic abundance (%)

L. crispatus
L. iners
L. roti
L. gasseri
L. vaginatis
D. vulgaris
L. crispatus
L. jensenii
L. gasseri
G. vaginalis

Streptococcus agalactiae
Finegoldia magna
B. vulgatus
B. bifidum
Atopobium sp
Others

Gardnerella vaginalis
Bifidobacterium
Pr
Enterococcus sp
Streptococcus anginosus
Candidate division TM7
Bacteroidales
Streptococcus iners
Bacteroides
e
e
Staphylococcus
P.

Corynebacterium
Finegoldia magna
Bifidobacterium
Streptococcus agalactiae
Peptostreptococcus
Anaerococcus
L. gasseri
L. jensenii
Drivers of unstability

Modeling the dependence of the log of Jensen-Shannon divergence rate of change (estimate of stability) on the menstrual time (normalized time) - Bayesian Markov Chain Monte Carlo methods using linear mixed effect models.

At any given time, >25% of women are in a non-lactobacillus dominated state.

- This state is associated with high Nugent scores and higher pH
- High Nugent score is associated with increased risk of sexually transmitted infection acquisition and transmission, including HIV, as well as preterm birth
- These women are “normal” and healthy, but at increased risk of STI or other adverse outcomes
- Community stability/dynamics (frequency and duration of CST IV) might represent risks to disease

  Low stability = low resilience = increased risk

Understand the molecular basis of this association between stability and susceptibility using omics’ technologies
Stability and the community genome

- Is there a correlation between the genomics content of some of the species and community stability?
  - Use metagenomics analysis of microbial communities
  - Develop a elaborated bioinformatic pipeline to establish community composition and assemble the genomes of community members
  - Perform comparative genome analysis with known genomes
Conclusions

- The types and abundance of microbes found in the vaginal vary over short time intervals in some women and in other do not change.

- Changes can often coincide with menses, but sometimes do not - Somewhat personalized. To some extend sexual activity tend to destabilize the communities.

- Genomic content of individual members of a community might predict community stability

- Community stability/dynamics might represent risks to disease

  Low stability = low resilience = increased risk
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