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Background The humoral response to *Treponema pallidum* (*T. pallidum*), which causes syphilis, is divided into 'non-specific' anti-lipid and specific anti-treponemal protein antibodies. A four-fold reduction in anti-lipid antibodies is used to diagnose cure, which can take six or more months.

Quantitative PCR (qPCR) can measure *T. pallidum* DNA copies in blood and ulcer samples. Bacteraemia is more common and of higher load in early disease.

We present a pilot study monitoring the early treatment response in patients with infectious syphilis by qPCR.

Methods Patients with symptomatic primary or secondary disease were admitted to hospital and following baseline sampling were treated with 2.4 M units of benzathine penicillin. Whole blood was collected into EDTA and Tempus RNA preservation tubes and the ulcer sampled using a philtre paper strip every four hours for *T. pallidum* DNA (*tpp047* gene) and RNA (*16S rRNA*) quantification. Sampling ended when two consecutive PCRs were negative. Standard serological follow-up was performed.

Results Three men were recruited (two secondary, one primary). All were homosexual and two were HIV-1 infected.

Blood DNA quantification and clearance: A mean peak-level of 1611(range 1212) *tpp047* copies/ml was detected and mean half-life for clearance (t½ clearance) was 7.89 hours (range 5.34). Blood RNA: Mean peak-level 8829(range 20366) *16SrRNA* copies/ml blood; (t½ clearance) 5.24 hours (range 0.78). Ulcer DNA 1.14×10^5 copies/strip and RNA 4.35×10^7 copies/strip with a t½ (clearance) of 1.67 and 3.76 hours, respectively. *T. pallidum* nucleic acids were undetectable in all samples after 56 hours.

All patients had serology consistent with disease stage at baseline and cure at one month.

Conclusions

- *T. pallidum* qPCR presents a novel and quick way of monitoring early syphilis treatment efficacy. Both DNA and RNA may be suitable targets to measure bacterial clearance from blood and ulcer exudates.
- Ulcers may be non-infectious as soon as 56 hours post-treatment.

005.3 MORE THAN MEETS THE EYE: A MOLECULAR PHYLOGENETIC ANALYSIS REVEALS IMPORTANCE OF NOVEL BACTERIA IN BACTERIAL VAGINOSIS (BV)

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Background BV is a highly prevalent dysbiotic condition associated with adverse reproductive and health outcomes in women. BV is marked by loss of certain lactobacilli and acquisition of complex communities of anaerobic bacteria. Gram stain is the gold standard for diagnosis, wherein the abundances of four bacterial morphotypes are assessed; Lactobacillus, Gardnerella, Bacteroides and Mobiluncus. Newly described uncultivated bacteria are highly specific for BV, but no studies have investigated the association between these bacteria and Gram stain morphotypes.

Methods We examined the association of bacteria detected by broad-range 16S-rRNA gene PCR/pyrosequencing with bacterial morphotypes detected in Gram stains from 220 women with and without BV. We also used species-specific quantitative PCR and fluorescence in situ hybridization (FISH) methods to document concentrations of two bacteria with curved rod morphologies: *Mobiluncus* species and the uncultivated BV-associated bacterium-1 (BVAB1).

Results We provide evidence that curved Gram-negative rods designated Mobiluncus morphotypes by Gram stain are more likely BVAB1. Rank abundance plots of vaginal bacteria in women with curved rods (Nugent 9–10) showed that BVAB1 was the dominant bacterium (26%), while relative abundance of *Mobiluncus* was only 0.2%. BVAB1 sequence reads were also associated with Mobiluncus morphotypes ($p = 7.4E-06$). Among women with Nugent scores 9–10, the mean concentration of BVAB1 DNA was 2-log units greater than *Mobiluncus* ($p < 0.001$). FISH analyses also revealed that among women with Nugent scores of 10, the mean number of BVAB1 cells was greater than *Mobiluncus* cells ($p < 0.001$). In addition, we noted that *Prevotella* and *Porphyromonas* spp. are significantly associated with the Bacteroides morphotype, whereas *Bacteroides* species are rare.

Conclusions Gram stain morphotype designations for BV need revision to account for novel vaginal bacteria. These findings have major implications for studies using Gram stains as a proxy to describe the vaginal microbiota.

005.4 ASSOCIATION BETWEEN CHLAMYDIA TRACHOMATIS GENITAL INFECTION AND THE VAGINAL MICROBIOME

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Background *Chlamydia trachomatis* (CT) genital infection is one of the most widespread sexually transmitted infections (STIs). It is thought that certain types of vaginal microbiota are better able to prevent STIs, however, very little is known about the relationships between species and genomic composition of the vaginal microbiome and risk of infection. Information about these can be used to advance prevention strategies for STIs. We sought to characterise the vaginal microbiome at the time of diagnosis and after treatment of CT infection.

Methods 101 women with CT genital infection were recruited to a longitudinal study in Baltimore, MD. Participants were treated at diagnosis and returned for clinical visits every 3 months for 9 months, at which times the vaginal microbiota was determined using 16S rRNA analysis. We further applied high-throughput metagenomics to characterise the genomic makeup of the vaginal microbiome.

Results The average age of participants was 19.3 (range 14–28) and 92% were African-American. During CT infection, comprehensive surveys of the vaginal microbiota demonstrated two distinct community state types characterised by: (1) a complex assemblage of strict anaerobes with low proportions of *Lactobacillus* spp. (60% of samples) or (2) a high relative abundance of *Lactobacillus iners*-dominated communities (37% of samples). Metagenomic evaluation suggested unique characteristics of the genomes of the dominant species in these communities (i.e., *L. iners, Gardnerella vaginalis*). *L. iners* appeared more genetically diverse than other *Lactobacillus* species.

Conclusions Complex communities of strict anaerobes with low proportions of *Lactobacillus* or specific *L. iners* genome types represented a hallmark of the chlamydia-infected state in this population. Some *L. iners* may be better suited to adapt to diverse environments while others may be contributing factors to an at-risk microbiome. In the era of personalised medicine, future work will enhance our ability to intervene and establish a protective vaginal microbiome.