by metagenomic sequencing (NovaSeq), and absolute abundance of key bacterial species was assessed by qPCR.

Results Immediately following treatment, there was a marked drop in the vaginal level of proinflammatory cytokines (IL-1a, IL-6), chemokines (IL-8, MIP-1b, MIP-3a) and a marker of epithelial barrier disruption (sE-cad; all $p < 0.01$). The vaginal microbiome was generally shifted from BV to community state types (CSTs) dominated by *L. iners* (49%) or *L. jensenii* (31%). However, these CST shifts were driven by a dramatic (7–12-fold) reduction in the absolute abundance of BV-associated Gram negative anaerobes (*Prevotella* spp., *Gardnerella vaginalis*, *Atopobium vaginae*, and *Megasphaera*; all $p < 0.001$), while there were minor or no increases in the absolute abundance of *L. jensenii* (2.9-fold increase, $p = 0.006$) or *L. iners* (0.6-fold reduction, $p = 0.245$).

Conclusions Standard BV therapy reduced inflammatory cytokines/chemokines and enhanced epithelial integrity in the short term. Despite a shift to CSTs dominated by lactobacilli, these immune changes were linked to reductions in the absolute abundance of BV-associated anaerobes rather than to increased *Lactobacillus* abundance.

003.3 INTRAVAGINAL LACTIC ACID GEL VERSUS ORAL METRONIDAZOLE FOR TREATING WOMEN WITH RECURRENT BACTERIAL VAGINOSIS: THE VITA RANDOMISED CONTROLLED TRIAL

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Background Bacterial vaginosis (BV) is a common, embarrassing and distressing condition, with serious comorbidities. The VITA trial assessed the effectiveness of intravaginal lactic acid gel versus oral metronidazole for treating recurrent BV.

Methods Open-label, multicentre, parallel group, randomised controlled trial. Women aged ≥ 16 years with history of BV within the past 2 years and current symptoms were recruited at one GP practice and 19 sexual health centres in the UK. Participants were randomised to 5ml intravaginal lactic acid gel or oral 400mg twice daily metronidazole tablets, for 7 days. The primary outcome was participant reported resolution of BV symptoms by the end of Week 2, with subsequent follow up over 6 months. Semi-structured interviews explored tolerability and acceptability of the study treatments.

Results 518 participants were randomised (259 to metronidazole, 259 to lactic acid gel). Primary outcome data were available for 204 (79%) participants allocated to metronidazole and 205 (79%) allocated to lactic acid gel. Resolution of BV at Week 2 occurred in 143 (70%) participants in the metronidazole arm versus 97 (47%) participants in the lactic acid gel arm (adjusted risk difference -23.2% (95% CI -32.3 to -14.0%). Symptom resolution with no subsequent recurrences over 6 months occurred in 23% of those who took metronidazole and 16% of those using lactic acid gel. Side effects were more common following metronidazole compared to lactic acid gel. Interviews revealed that many women preferred lactic acid gel despite perceiving antibiotics to be more effective.

Conclusion Metronidazole is more effective than lactic acid gel in achieving short term resolution of BV symptoms but recurrence is common following both treatments. Lactic acid is associated with fewer reported side effects and is preferred by many women despite its lower efficacy.

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Trial Registration ISRCTN14161293

003.4 THE VAGINAL MICROBIOME AND DEVELOPMENT OF PELVIC INFLAMMATORY DISEASE: A PROSPECTIVE STUDY USING 16S RRNA LONG-READ SEQUENCING

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Background A low diversity vaginal microbiome dominated by *Lactobacilli*, may be associated with lower risk of pelvic inflammatory disease (PID). However, disruption to this balance through an increase in diversity, particularly dominated by *Gardnerella vaginalis*, may lead to increased susceptibility. Not all *Lactobacilli* are equally protective, species that produce higher proportions of D(-) isomer lactic acid (*L. crispatus*, *L. jensenii*, and *L. gasseri*) have proven protection against *Chlamydia trachomatis* (CT) infection, an important cause of PID, however *Lactobacillus iners*, which produces predominantly L (+) isomer lactic acid, may be less protective. We investigated the relationship between 'baseline' microbiome and the subsequent risk of acquiring PID, using stored vaginal samples from participants in the Prevention of Pelvic Infection chlamydia screening trials.

Methods 16S rRNA long-read nanopore sequencing was carried out on vaginal samples from 37 women who subsequently developed clinically defined PID during 12 months of follow-up, and 111 frequency-matched controls who did not. Samples were matched on age, ethnicity, CT status at baseline and number of sexual partners.

Results Three main taxonomic clusters were identified which were dominated by *L. iners*, *L. crispatus*, and *G. vaginalis*. Microbiomes dominated by *G. vaginalis* were associated with black ethnicity ($p = 0.002$), age ≥ 20 ($p = 0.048$) and bacterial vaginosis ($p < 0.0001$), but not with subsequent development of PID or CT infection at baseline. Lower rates of D(-) isomer lactic acid producing *Lactobacilli* were not associated with PID ($p = 0.53$) or concurrent CT ($p = 0.38$) infection, but were associated with bacterial vaginosis, as expected ($p < 0.0001$).

Conclusion We found no association between baseline vaginal microbiome composition and subsequent development of PID, nor between quantities of D(-) isomer lactic acid producing *Lactobacilli* and concurrent CT infection.

003.5 PID TREATMENT AND MYCOPLASMA GENITALIUM (MG): THE IMPACT OF VAGINAL DYSBIOSIS

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Background In a recent PID treatment trial, MG was recovered significantly less from women treated with a regimen that included metronidazole. Our objective was to determine if cervical MG at randomization and following treatment correlated with vaginal dysbiosis.

Methods This is a secondary analysis of the ACE Trial, a randomized trial comparing single dose ceftriaxone and two weeks of doxycycline, with either metronidazole 500 mg PO BID or placebo for 2 weeks. At enrollment and at 30 days following treatment, vaginal samples were evaluated for BV by Nugent criteria, and quantitative PCR was performed for anaerobic organisms. Endocervical swabs were tested for MG by NAAT.

Results At randomization, MG infection was detected in 41/233 (18%) women, but detection at follow-up differed for those who received metronidazole [4/94 (4%)] vs placebo [14/99 (14%), $p < 0.05$]. Women having MG at baseline had higher concentrations of four microbes (*Gardnerella vaginalis*, *Atopobium vaginae*, *Megasphaera lornae*, *Prevotella timonensis*) compared to women without cervical MG ($P < 0.05$ for each), and each of these microorganisms was associated with cervical MG after treatment ($P < 0.05$ for each). Although BV by Nugent criteria was not significantly associated with MG at randomization, women without BV following PID treatment were less likely to have MG infection than women with BV (4% vs 18%, $p < 0.01$).

Conclusion MG infection following PID is uncommon among women treated with metronidazole along with ceftriaxone and doxycycline, despite limited activity of this regimen against MG. MG infection was associated with vaginal dysbiosis, as defined by higher densities of BV-associated bacteria. These results suggest that a PID regimen with oral metronidazole and eradication of BV pathogens might be key to eliminating MG in women. Whether women diagnosed with PID should be tested for MG and treated with targeted therapy if positive remains uncertain.

003.6 VAGINAL WASHING BEHAVIOR AND FECUNDABILITY IN KENYAN WOMEN PLANNING PREGNANCIES

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Background Vaginal washing is associated with acquisition of HIV, sexually transmitted infections, and bacterial vaginosis (BV). One study of US women found that vaginal washing was associated with a 30% reduction in fecundability, the perimenstrual cycle probability of pregnancy. We examined the association between vaginal washing and fecundability among Kenyan women.

Methods HIV-negative Kenyan women who were trying to conceive and reported no history of infertility care-seeking were followed prospectively for incident pregnancy for up to six months. At monthly visits, participants reported the first day of last menstrual period, sexual behavior, vaginal washing

behavior, underwent pregnancy testing, and provided vaginal swabs for detection of BV by Gram stain (Nugent score ≥ 7). Discrete time proportional probabilities models were used to estimate fecundability ratios (FR) and 95% confidence intervals (CI) comparing menstrual cycles when women reported vaginal washing to menstrual cycles when no vaginal washing was reported.

Results 458 women contributed 1,376 menstrual cycles and 255 pregnancies. At enrollment, 7.5% ($n=34/454$) women had Chlamydia trachomatis detected, 0.7% ($n=3/454$) had Neisseria gonorrhoeae, 0.9% ($n=4/453$) had Trichomonas vaginalis, 35.8% ($n=164/458$) had BV, and 10% ($n=46/458$) reported abnormal vaginal discharge. A third (35.4%, $n=162/458$) of participants reported vaginal washing at enrollment with the majority using water only (72.8%, 118/162). After adjustment for age, frequency of condomless sex, and study site, vaginal washing in the prior four weeks was associated with a significant 29% lower fecundability (adjusted FR (aFR) 0.71, 95%CI 0.53–0.94), which did not change after further adjustment for BV at the visit prior to each pregnancy test (aFR 0.71, 95% CI 0.54–0.97).

Conclusions Periconceptual vaginal washing may reduce fecundability. Potential mechanisms include vaginal washing-associated changes in the vaginal microbiota, inflammation, disruption of cervical mucus, and effects on sperm function. Vaginal washing has no known health benefits, and cessation may improve women's likelihood of conceiving.

COVID – epidemiology & health services impacts

004.1 THE IMPACT OF THE CORONAVIRUS DISEASE (COVID-19) ON THE HEALTH AND SOCIAL NEEDS OF SEX WORKERS IN SINGAPORE

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Background We evaluated the impact of the Coronavirus Disease (COVID-19) on the sex work industry, and assessed how it has impacted the health and social conditions of sex workers in Singapore.

Methods We conducted a sequential exploratory mixed methods study amidst the COVID-19 pandemic from April to October 2020, including in-depth interviews with 24 stakeholders from the sex work industry and surveyor-administered structured surveys in the field with 171 sex workers. The in-depth interviews allowed the team to qualitatively explore and generate themes around how COVID-19 had impacted sex workers in Singapore, and informed the design of the quantitative surveyor-administered survey questionnaire. Qualitative data were analyzed through framework analysis while survey data were analyzed through descriptive statistics, and multi-variable Poisson regression models.

Results COVID-19 had a substantial impact on sex workers' income, while the illegality of sex work, stigma, and the lack