Methods MtrR was ectopically expressed in HO41 (named as SC4) and confirmed to be functional by western blot and qRT-PCR analyses. HO41 and SC4 were compared for their susceptibility to antibiotics in laboratory media and in the presence of ME180 cervical epithelial with or without IPTG induction.

Results In both laboratory media and in ME180 cell culture, we found that expression of MtrR in SC4 (HO41 *mtrR*+) decreased *mtrCDE* gene expression and increased gonococcal susceptibility to beta-lactam antibiotics. Importantly, MtrR-mediated repression of *mtrCDE* decreased the MIC of penicillin to a level below the MIC breakpoint recommended clinical treatment dose.

Conclusion We demonstrate the MtrR-mediated dampening of mtrCDE can greatly increase gonococcal susceptibility to penicillin. Thus, novel adjunctive therapeutics that decrease levels of MtrCDE may allow for the return of penicillin as an option for treating otherwise resistant strains of gonococci.

P1.12 ROLE OF VAGINAL LACTOBACILLI IN COUNTERACTING *CHLAMYDIA TRACHOMATIS* INFECTIVITY IN AN IN VITRO MODEL

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Introduction Lactobacilli play a fundamental role in maintaining the ecological equilibrium of the vaginal niche, preventing the overgrowth of endogenous microorganisms and impeding the colonisation of pathogens. Although many studies have focused on the mechanisms displayed by lactobacilli in counteracting several urogenital pathogens, a few data are available on the interaction between lactobacilli and *Chlamydia trachomatis* (CT). The aim of this study was therefore to assess the *in vitro* activity of different vaginal *Lactobacillus* strains against CT infectivity, investigating two different fractions of bacteria (cell pellets and cell-free supernatants), by three different mechanisms of action (competition, exclusion and displacement).

Methods A total of 17 *Lactobacillus* strains, isolated from vaginal swabs of healthy premenopausal women and belonging to *L. crispatus* (BC1; BC3-BC8), *L. gasseri* (BC9-BC14), *L. vaginalis* (BC16-BC17) and *L. plantarum* (BC18-19) species, were included in the study. The capacity of lactobacilli cell pellets (CP) and cell-free supernatants (CFS) to interfere with CT adhesion and entry in HeLa epithelial cells was evaluated, by means of competition, exclusion and displacement mechanisms. In particular, lactobacilli fractions corresponding to 5×10^7 colony forming units (CFU) were incubated with 5×10^3 CT elementary bodies (EBs) of strain GO/86, (serotype D), following different timelines. CT infection was evaluated by counting chlamydia inclusion forming units (IFUs) by direct immunofluorescence. Moreover, on the basis of CT infectivity interference results, 5 lactobacilli were selected for dose-effect assays and the same experiments were repeated, using CP or CFS fractions with 5×10^6 and 5×10^5 lactobacilli cells, in order to verify if a dose-dependent activity was present. Finally, the capacity of *Lactobacillus* strains to adhere to HeLa cells was assessed as well: results were read at light-microscopy and HeLa cells were scored for the presence and number of lactobacilli attached.

Results We found that lactobacilli cell pellets were the most active fraction in counteracting CT infectivity, particularly by means of an exclusion strategy, and that *L. crispatus* was the most effective species, even though a strain-specific activity was detected. Moreover, the anti-chlamydial activity was not correlated with the level of lactobacilli adhesion on epithelial cells and it was significantly maintained with low numbers of lactobacilli, although in presence of a dose-response effect.

Conclusion We identified specific vaginal *Lactobacillus* strains (*L. crispatus* BC4, *L. crispatus* BC5, *L. crispatus* BC7, *L. gasseri* BC14 and *L. plantarum* BC19) able to interfere with CT EBs adhesion and entry in epithelial cells and we were able to shed light on the mechanisms displayed by lactobacilli in counteracting CT infectivity. A major potential application lies on the use of these *Lactobacillus* strains as probiotics for the prophylaxis and/or adjuvant therapy of CT infections.

P1.13 VAGINAL MICROBIOME SIGNATURES IN *CHLAMYDIA TRACHOMATIS* INFECTED WOMEN

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Introduction In healthy women, lactobacilli play a crucial role in maintaining the microbial homeostasis of the vaginal niche. In case of bacterial vaginosis (BV), a condition characterised by a depletion of lactobacilli and an increasing number of anaerobes, a higher risk of urogenital and sexually transmitted infections (STIs) is reported. The vaginal environment of healthy and BV-positive women have been extensively studied, leading to the identification of the microbial species dominating these opposite conditions and to the description of specific metabolic profiles. Besides that, less is known about the vaginal microbiome in case of STIs, as *Chlamydia trachomatis* (CT) infections. The aim of this study was to analyse the composition of the endogenous microbiota and the metabolic signatures of the vaginal niche in 3 different conditions: healthy, BV and CT infections.

Methods From July 2016, all the pre-menopausal women attending the STI Outpatients Clinic of Sant'Orsola-Malpighi Hospital in Bologna (Italy) and meeting one of the following criteria were enrolled: presence of vaginal symptoms or presence of risk factors for CT infection. Patients with vaginal candidiasis were excluded. For all the patients, a vaginal swab was collected for molecular CT detection (Versant CT/GC DNA 1.0 Assay; Siemens), whereas Amsel criteria were used for BV assessment. Moreover, for each woman, an additional vaginal swab stored in saline was collected and centrifuged. Cell pellets were examined with a DNA-microarray platform including 17 probe sets specific for the most representative vaginal bacterial groups and with a quantitative real-time PCR targeting 16s rRNA gene of *Gardnerella vaginalis* (GV). Cell-free supernatants were used for metabolomic analysis by means of ^{1}H-NMR spectroscopy. NMR spectra were recorded with an AVANCE spectrometer (Bruker). Similarities among microbial and metabolic profiles of samples were investigated by means of a principal component analysis (PCA). Differences in GV DNA loads and metabolites concentrations were

analysed by ANOVA test. The study was approved by the Hospital Ethical Committee.

Results Among all the women enrolled, 25 were considered healthy, 18 received a diagnosis of BV and 22 were positive for CT. PCA revealed that the vaginal microbiome of healthy and BV-subjects were clearly distinct and that CT-positive women were more similar to healthy women rather than to BV-positives, both for microbial composition and for metabolic profile. The mean GV DNA load was significantly different between the groups (p=0.03): healthy and CT positive women showed similar and lower mean loads compared to BV group. At a metabolic level, significantly higher concentrations of formate, ethanolamine and methylamine were found in BV-patients, while tryptophan and lactate were more present in healthy and CT-positive women.

Conclusion Specific microbial and metabolic signatures characterise different clinical conditions of the vaginal tract. In this context, CT-positive women are definitely more similar to healthy than BV-subjects.

P1.14 EVALUATION OF THE APTIMA ASSAYS FOR THE DETECTION OF BACTERIAL SEXUALLY TRANSMITTED INFECTIONS IN A SELECTED POPULATION OF WOMEN

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Introduction: *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (NG) and *Mycoplasma genitalium* (MG) represent the most common agents of bacterial sexually transmitted infections (STIs), worldwide. In women, uro-genital infections caused by these microorganisms are often asymptomatic and, left untreated, can lead to several sequelae. Nucleic acid amplification techniques (NAATs) have become the reference methods for the diagnosis, thanks to the suitability for different specimens and the outstanding sensitivity and specificity. The aim of this study was to assess the performance of Aptima Assays for CT, NG and MG detection in a group of selected women, by a head-to-head comparison with other NAATs. Moreover, an evaluation about the suitability for the Aptima assays with one of the most used swab collection device (E-Swab; Copan) was carried out.

Methods Routinely, all the women attending the STI Outpatients Clinic of Sant'Orsola-Malpighi Hospital of Bologna (Italy) complaining of genital STI-related symptoms or reporting unsafe intercourse, are managed as follows. After a clinical visit, a sample of first-void urines and a vaginal swab collected in E-swab, are obtained for CT, NG and MG detection. A duplex real-time PCR (Versant CT/GC DNA 1.0 assay; Siemens) is used for CT and NG detection, while, MG presence is investigated by a home-made PCR, starting from the remaining eluate of Versant PCR plate. From January 2016, a total of 100 patients were selected and their samples were also tested with Aptima assays. Previously frozen samples were thawed and transferred to the suitable collection devices for Aptima assays: in particular, 2 ml of urines and 100 µl of vaginal E-swab were used. All the specimens were processed by Aptima Combo2® for CT and NG detection and by the Aptima® *Mycoplasma genitalium* assay for MG infection diagnosis. These assays were run on Panther system (Hologic). A comparison between the different molecular methods, stratified by type of sample and microorganism, was conducted.

Results In the group of 100 women selected, 25 patients were positive for CT, 4 for NG and 6 for MG. One case of CT-NG and two cases of CT-MG co-infections were found. Interestingly, more than 50% of CT-positive women were completely asymptomatic. By the routine tests, all positive cases were simultaneously found both on the urine sample and on the vaginal swab, except for 3 CT, 1 NG and 1 MG infections, detected only on the vaginal swab. A complete concordance with Aptima assays, both for the type of sample and microorganism was found, with only one discordant result (a CT case detected by Versant on urines and vaginal swab, found by Aptima only on urines). Any interference due to the different liquid components of E-Swab was excluded.

Conclusion Given the outstanding performance, Aptima assays can represent an excellent choice for CT, NG and MG molecular detection. Moreover, it is noteworthy that Aptima assays allow testing of specimens collected by E-Swab, enabling the possibility to use the same sample for both NG molecular detection and culture.

P1.15 *TRICHOMONAS VAGINALIS* IN HUMAN IMMUNODEFICIENCY VIRUS-INFECTED PREGNANT WOMEN: PREVALENCE, DETECTION, AND APPLICATION OF PCR CYCLE-THRESHOLD VALUES

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Introduction: *Trichomonas vaginalis* (TV) is a sexually transmitted infection (STI) associated with increased transmission of human immunodeficiency virus (HIV) and significant adverse birth outcomes. Although culture is often used in the diagnosis of TV, molecular diagnosis is rapid, accurate, and data-rich.

Methods Women were recruited from two clinics in South Africa as part of a study assessing point-of-care polymerase chain reaction (PCR) diagnosis and treatment of three STIs. HIV-infected pregnant women were screened for TV using the Xpert TV (Cepheid, Sunnyvale, CA). Each woman who tested TV PCR-positive provided an additional sample for culture (InPouch TV, Biomed, San Jose, CA). We compared TV detection between PCR and culture, and used non-parametric statistics to compare cycle threshold (Ct) values among culture results and treatment outcomes.

Results By December 31st, 2016, 200 women were enrolled and 52 (26%) tested TV PCR-positive. Baseline cultures were obtained from 41 (79%) of the TV PCR-positive women, and 22 (54%) were culture-positive. The median baseline Ct of the TV PCR-positive/culture-positive group was 24.0 (IQR: 5.1) vs. 38.0 (IQR: 3.8) among those TV PCR-positive/culture-negative (p<0.05). Forty-two women returned for a 3 week test-of-cure (ToC), and 10 (24%) were still TV PCR-positive. Of the women who remained TV PCR-positive at