

P05.13 PREVALENCE AND ANATOMICAL DISTRIBUTION OF *MYCOPLASMA GENITALIUM* MACROLIDE RESISTANCE MARKERS FROM SUBJECTS ENROLLED IN A MULTI-CENTRE US CLINICAL STUDY

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Introduction This study evaluated the prevalence and anatomical distribution of *Mycoplasma genitalium* (Mgen) 23s rRNA mutations (23s-MTs) conferring macrolide resistance among male and female subjects enrolled in a prospective multi-site US clinical study.

Methods Specimens obtained from symptomatic and asymptomatic men and women enrolled from 7 diverse US clinical sites, including obstetrics and gynaecology, family planning, public health, and sexually transmitted disease clinics, were tested using a research TMA assay for Mgen (Hologic, Inc.) on the DTS System or TIGRIS DTS System. Samples positive for Mgen by TMA were evaluated by nested PCR or RT-PCR and Sanger sequencing of Mgen 23S rRNA to identify the presence of macrolide resistance mutations at position 2058 (A2058G, A2058C, A2058T) or position 2059 (A2059G).

Results Of 50 male subjects with Mgen 23s rRNA sequence results, 21 (42%) contained 23s-MTs. Slightly more 23s-MTs were found in urethral swabs vs male urine samples (44.8% vs 36.7%, respectively). For female subjects, 65 of 128 (50.8%) harboured 23s-MTs, with higher prevalence found in vaginal swab samples (50.2%) compared to urine (46%), Thinprep liquid Pap (41.7%), and endocervical swabs (37.8%). Sequencing of samples collected from anatomically distinct female urogenital sites (vagina, cervix, urine) revealed 18 of 35 (51.4%) subjects had a unique complement of Mgen 23s-MT and/or wild-type sequences at each anatomic site. For male subjects with both urine and urethral swab samples, 3 of 9 (33.3%) subjects had unique Mgen 23s-MT/WT sequences in each sample type.

Conclusion Mgen strains harbouring 23s rRNA-mediated macrolide resistance phenotypes were highly prevalent in this US cohort of male and female subjects. Detection of different macrolide-resistant Mgen strains in samples collected from different anatomical locations suggests that previous estimates for resistance rates that relied on a single anatomical site sample collection may have underestimated the extent of Mgen macrolide resistance in the population.

Disclosure of interest statement D Getman, M O'Donnell, and A Jiang are scientists employed by Hologic. S Cohen is a student at Occidental College and a summer intern at Hologic.

P05.14 MINIMUM INHIBITORY CONCENTRATIONS OF METRONIDAZOLE AND TINIDAZOLE AGAINST *TRICHOMONAS VAGINALIS*

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Introduction Syndromic management is used to control sexually transmitted diseases in South Africa. *Trichomonas vaginalis* causes trichomoniasis which results in vaginal discharge in symptomatic patients. This is treated with metronidazole which is included in the syndromic management antimicrobial regime. In order for this regime to be effective, the organisms causing each syndrome and their antimicrobial susceptibility profile need to

be evaluated periodically to ensure that the most appropriate antimicrobial agents are included.

Methods Women 18 years and older presenting with vaginal discharge were recruited from two different clinics of KwaZulu-Natal province in South Africa. Vaginal specimens were collected using a Dacron swab and cultured in modified Diamonds medium. The minimum inhibitory concentrations (MICs) of *T. vaginalis* to metronidazole and tinidazole were determined in 94 positive clinical isolates using a micro-broth dilution method. Briefly trichomonads were added to Diamonds media containing two-fold dilutions (16 to 0.25 mg/L) of metronidazole or tinidazole and incubated anaerobically for 72 h. The lowest concentration at which no motile trichomonads were visualised under an inverted phase contrast microscope was considered the MIC. *Propionibacterium acnes* and *Bacteroides fragilis* were used as the resistant and sensitive controls respectively. MIC \leq 1 mg/L was considered sensitive, MIC \geq 4 mg/L was considered resistant; MIC between 1 mg/L and 4 mg/L was considered intermediate. The MIC of any isolate in the resistant range was repeated to confirm results.

Results Of the 94 isolates, 17 had an MIC \geq 4 mg/L indicating *in vitro* resistance to metronidazole while 2 isolates had an MIC \geq 4 mg/L for tinidazole. Thirty-five and 33 isolates had an MIC of 2 mg/L (intermediate) for metronidazole and tinidazole respectively.

Conclusion High MIC of *T. vaginalis* to metronidazole is a public health concern however more research is needed to correlate *in vitro* resistance with clinical failure.

Disclosure of interest statement The authors have no conflict of interest to declare. No pharmaceutical grants were received in the development of this study.

P05.15 *UREAPLASMA* SPP. ISOLATED FROM GENITAL SAMPLES IN SWITZERLAND: SUSCEPTIBILITY PATTERNS, RESISTANCE GENES, AND SEQUENCE TYPE DISTRIBUTION

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Background Antibiotic resistance in *U. urealyticum* (UUA), *U. parvum* (UPA) and *M. hominis* (MH) poses an increasing issue. However, data regarding antibiotic susceptibility is limited to several countries, whereas information about clonality is available only from China.

Methods We analysed 140 genital samples collected in two laboratories from unique patients in Bern during 2014. Identification and antimicrobial susceptibility tests were obtained using the mycoplasma IST 2 kit (bioMérieux) and sequencing of 16S rDNA. Clonality was analysed with multilocus sequence typing (MLST) and expanded MLST (eMLST), whereas quinolone and macrolide resistance were studied by sequencing *gyrA/B*, *parC/E*, as well as genes encoding 23S rRNA and L4/22 ribosomal proteins.

Results One-hundred-three samples (74%) were confirmed being positive for UUA/UPA, whereas 21 (15%) were positive for both UUA/UPA and MH. Non-susceptibility was highest to ciprofloxacin (19.4%) and ofloxacin (9.7%), whereas low rates were observed for clarithromycin (4.8%), erythromycin (1.9%), azithromycin and tetracycline (both <1%). Various Sequence Types

(STs) previously reported in China (ST1, ST2, ST4, ST9, ST22, ST47), but also eight novel lineages, were detected. Only some quinolone-resistant isolates had amino acid substitutions in ParC (Ser83Leu in UPA) and ParE (Val417Thr in UPA and the novel Thr417Val in UUA), whereas the mechanism (s) for the remaining strains remains unclear. Although several isolates were non-susceptible to macrolides, mutations in 23S rRNA or substitutions in L4/L22 were not detected.

Conclusion This is the first study analysing susceptibility of *Ureaplasma* spp. isolates detected in Switzerland and the clonal distribution outside China. Resistance rates are low compared to other surrounding countries, but the empirical use of quinolones is compromised. We hypothesise that some hyperepidemic STs (e.g., ST4) spread worldwide via sexual intercourse. Large combined microbiological and clinical studies should address this important aspect.

P06 - Genital microbiome

P06.01 WOMEN OF DUTCH ETHNIC ORIGIN HAVE LOWER PREVALENCE OF VAGINAL MICROBIOME DYSBIOSIS THAN WOMEN OF OTHER ETHNIC ORIGIN RESIDING IN AMSTERDAM

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Introduction American women of African or Hispanic ancestry have increased risk of vaginal microbiome dysbiosis compared to women of European or Asian ancestry. However, the association between vaginal microbiome composition and ethnicity within Europe is largely unknown. We investigated this association in Amsterdam, The Netherlands.

Methods Non-pregnant women (18–34 years, n = 564) representing six ethnic origins (Dutch, South-Asian/Indonesian Surinamese, African Surinamese, Ghanaian, Turkish, and Moroccan) were cross-sectionally selected from the ongoing HELIUS multi-ethnic cohort study in Amsterdam for vaginal microbiome analysis. Extracted DNA from self-sampled vaginal swabs was sequenced targeting the V3V4 region of the 16S rRNA gene and using the Illumina MiSeq platform, and sequence reads were clustered using hierarchical clustering.

Results Clustering of 502/564 samples with sufficient read counts resulted in microbiome clusters dominated by *Lactobacillus crispatus* (n = 120), *L. iners* (n = 168), *L. jensenii* (n = 8), *L. gasseri* (n = 10), *Streptococcus agalactiae* (n = 8), *Bifidobacteriaceae/Bifidobacterium* spp. (n = 10), *Gardnerella vaginalis* (n = 78), and a mixture of anaerobes (n = 100), respectively. Microbiome composition was significantly associated with ethnic origin (P = 0.002). Women of Dutch ethnic origin had the highest prevalence of *L. crispatus*-dominated microbiome (40% vs 16–26% in the other ethnic groups), the lowest prevalence of *L. iners*-dominated microbiome (28% vs 31–39% in the other groups), and the lowest prevalence of clusters dominated by *G. vaginalis* or a mixture of anaerobes (25% vs 30–45% in other groups). Turkish women and South-Asian/Indonesian Surinamese

women had the highest prevalence of *L. iners*-dominated microbiome (38% and 39%, respectively), and women from African descent (African Surinamese and Ghanaian women) the highest prevalence of clusters dominated by *G. vaginalis* or a mixture of anaerobes (48% and 44%, respectively).

Conclusion This large multi-ethnic study shows that dysbiotic vaginal microbiome compositions are significantly increased in women of non-Dutch ethnic origin. Therefore, these women may be at increased risk of STI acquisition and adverse reproductive health outcomes.

Disclosure of interest statement The HELIUS study is funded by the Academic Medical Centre Amsterdam, the Public Health Service of Amsterdam, the Dutch Heart Foundation (project number 2010T084), the Netherlands Organisation for Health Research and Development (ZonMw; project number 200500003), and the European Union (FP-7; project number 278901). The vaginal microbiome analyses were funded by the Aids Fonds Netherlands (project number 201102). The authors declare no conflicts of interest.

P06.02 LONGITUDINAL CERVICOVAGINAL MICROBIOME MEASUREMENTS OF WOMEN BEFORE AND AFTER HIV-SEROCONVERSION

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Introduction Bacterial vaginosis (BV) by Nugent scoring is associated with enhanced acquisition and cervicovaginal shedding of HIV, but longitudinal molecular studies of these relationships are scarce.

Methods HIV-negative (n = 397) female sex workers in Kigali, Rwanda, were followed for two years. Demographic, behavioural, clinical, HIV, sexually transmitted infection, and cervicovaginal microbiota data were collected at regular intervals. The cervicovaginal microbiota were characterised by Nugent scoring, Amsel criteria (pH >4.5, a positive whiff test and presence of >20% clue cells on wet mount; two or three criteria indicated BV diagnosis), and phylogenetic 16S DNA microarray.

Results During follow-up, 19 women seroconverted for HIV. The associations between BV by Nugent or Amsel criteria and subsequent HIV seroconversion did not reach statistical significance (aOR = 1.56 (95% CI 0.51–4.77) and aOR = 4.85 (95% CI 0.59–39.90), respectively). For 10/19 women, phylogenetic microbiome composition was available before and after seroconversion, with a median of 324 days (range 42–386) before and 196 days (range 121–492) after seroconversion. Before seroconversion, none of the women had a *L. crispatus*-dominated microbiome, four had a *L. iners*-dominated microbiome, four a moderately dysbiotic microbiome and two severe dysbiosis. The microbiome composition of five women remained stable before and after seroconversion, four shifted to (more severe) dysbiosis, and one shifted from dysbiosis to a *L. iners*-dominated microbiome. After seroconversion, phylogenetic microbiome composition was available for all 19 women. 26% had a *L. iners*-dominated microbiome, 32% moderate dysbiosis, and 42% severe dysbiosis.

Conclusion In this population of high risk women, the association between BV and subsequent HIV seroconversion did not reach statistical significance, but statistical power was limited.