**P3.270** ANTIMICROBIAL SUSCEPTIBILITY AND MOLECULAR CHARACTERIZATION OF NEISSERIA GONORROEA Strains FROM SLOVENIA 2008–2012

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**Objectives** Antimicrobial resistance in Neisseria gonorrhoeae is a major public health problem worldwide. Slovenia is among the countries with highest prevalence of decreased susceptibility and resistance to extended-spectrum cephalosporins (ESCs) in Europe. Herein, the phenotypic and molecular characterization of Slovenian N. gonorrhoeae strains from 2008–2012 is presented.

**Methods** N. gonorrhoeae isolates cultured 2008–2012 in Slovenia (n = 138) were examined for antimicrobial susceptibility with Etest methodology for 8 antimicrobials (cefixime, ceftriaxone, penicillin, ciprofloxacin, azithromycin, tetracycline, gentamicin and spectinomycin). Furthermore, all isolates were investigated with Neisseria gonorrhoeae multi-antigen sequence typing (NG-MAST) for molecular epidemiology, and sequencing of major ESC resistance determinants; penA, mtrR and penB.

**Results** The overall prevalence of resistance and decreased susceptibility to cefixime and ceftriaxone (MICc ≤ 0.125 mg/L) was 16% and 7%, respectively. The resistance and decreased susceptibility showed an epidemic peak during 2009–2011 when it reached 28% for cefixime and 12% for ceftriaxone, however, the prevalence subsequently decreased to 6% and 4%, respectively, in the year 2012. NG-MAST sequence types 1407 (13% of all isolates), 21 (8%) and 225 (6%) were the most common STs during 2008–2012. ST1407, previously stated as an internationally spread successful clone with resistance or decreased susceptibility to ESCs, was most prevalent in 2009 (45%). However, the ST1407 prevalence declined in 2010 (15%) and 2011 (11%) and the clone was rare in 2012 (4%). Instead, in 2012 the ESC susceptible ST21 was the predominant ST (21%). During 2008–2012, a penA mosaic allele (mainly penA XXXIV, associated with ST1407) was detected in 25% of isolates.

**Conclusion** The prevalence of gonococcal resistance to ESC in Slovenia has fluctuated, which is mainly due to the longitudinal prevalence of a few gonococcal clones. Fortunately, some ESC susceptible clones, such as ST21, now appear to replace the main ESC resistant clone ST1407, a replacement that is indicated in several European countries.

**P3.271** IDENTICAL MULTILOCUS SEQUENCE TYING (MLST) ANALYSIS IN SEQUENTIAL SAMPLES FROM PATIENTS WITH PHARYNGEAL CHLAMYDIA INFECTIONS

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**Introduction** Pharyngeal Chlamydia trachomatis (PtC) must persist to contribute to ongoing transmission. In a retrospective study, we examined MLST-types of PtC in patients who had a positive pharyngeal swab on two visits, and had not been treated for this infection at first visit.

**Methods** From 1/1/2008 to 14/7/2010, pharyngeal swabs from patients at risk for pharyngeal gonorrhoea were tested with the AC2 (Hologic-GenProbe) test. Since at that time PtC detection was not considered to represent an infection, PtC results were not reported and patients were not treated, unless they had a chlamydial infection at another anatomic site. We looked for patients who had a positive PtC test on two different occasions with an interval of at least 3 weeks. For inclusion in the study, patients were required to have no Chlamydia infections at other anatomic locations at first visit and therefore received no treatment. PtC typing was done by MLST on stored specimens.

**Results** Sixteen patients could be included and paired pharyngeal samples from four of those patients contained enough DNA for MLST analysis. The intervals between the two visits were 112, 168, 207 and 268 days, respectively. In all four patients MLST types of both pharyngeal samples were completely identical. Patients were two women and two men who had sex with men (MSM). At second visit one woman and one MSM reported commercial sex work and had 30 and 150 sexual partners in the last 6 months, respectively. The second woman reported sex with two known persons and the second MSM reported sex with 15 known persons. None reported sex with a steady partner.

**Conclusion** Our findings of identical MLST types are consistent with persistent PtC infection for a period of 5–9 months, although repetitive exposure to untreated partners with identical C. trachomatis strains cannot be excluded.

**P3.272** PREVALENCE OF CHLAMYDIA TRACHOMATIS, NEISSERIA GONORROEA AND UREAPLASMA UREA LYCITICUM IN PREGNANT WOMEN OF SABZEVAR - IRAN

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**Background** One approach to eliminating sexually transmitted infections (STIs) in a community is to screen high-risk persons, followed by the treatment and education of people who test positive. We examined a Triplex PCR assay to detect urine samples in pregnant women. Major goal of this study was to determine the prevalence of Chlamydia trachomatis (Ct.), Neisseria gonorrhoeae (Ng.) and Ureaplasma urealyticum (Uu.) and to apply a Triplex PCR techniques to detect 3 pathogens in one specimens.

**Methods** A cross-sectional study was conducted among 399 pregnant adolescents women. All participants received prenatal care and delivered at urban hospital in Sabzevar- Iran. A socio-demographic questionnaire was completed. A Triplex PCR with universal primers was developed for screening of subjects women in this study. Descriptive and univariate analyses were performed to describe disease prevalence.

**Results** Of pregnant adolescents, 15.28% were diagnosed with either Ct., Ng. or Uu. infection. The prevalence of Ct. was 12.28%, and that of Ng 1.25% and finally 19.54% of pregnant had Uu. infection.

In univariate analysis, Ct. was associated with having had any level of education (P < 0.05), abortion (P < 0.05), and Uu. was associated with PTD (P < 0.05). Multivariate analysis did not show any significant association.

**Conclusions** Compared with available data, a decline of STIs prevalence was observed in our setting. This might be the result of
community-based education programmes focusing on changes to sexual behaviour. However, STIs rates are still high, and the problem needs more concrete and sustained efforts for its control. Screening for Ct., Ng. and Uu is recommended during pregnancy. Based on our finding in this study, the overall high incidence of Ct. and Uu support screening recommendations for pregnant women in Sazevar- Iran.

**P3.273** MOLECULAR TYPING AND DETECTION OF MACROLINE RESISTANCE MUTATIONS IN T. PALLIDUM STRAINS FROM CALI, COLOMBIA

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**Background** Cali is a syphilis-endemic area, with a very high prevalence of gestational syphilis (14.7/1000 live births) and congenital syphilis (7.7/1000 live births). Molecular typing techniques are useful for studying bacterial strain diversity, molecular epidemiologic patterns and antimicrobial resistance patterns. The study objective was to determine the *T. pallidum* (*Tp*) strain diversity and analyse for the mutations associated with macrolide-resistance in this syphilis-endemic region.

**Methods** 19 secondary syphilis (SS) patients, 15 (RPR+, FTA-ABS+, HIV-) and 4 (RPR+, FTA-ABS+, HIV+) were enrolled in the study. *Tp* DNA was extracted from syphilis skin lesions and quantified by real-time-qPCR targeting *polA* gene. Molecular typing was performed using established typing and subtyping methods (*arp*, *tp*, *tp0548*) and strains were analysed for *A2058G* and *A2059G* mutations within the 23S rRNA gene. Strain diversity in Cali strains (*arp* and *tp*) was compared with other published *Tp* molecular studies using the Shannon index.

**Results** 14/19 SS patients were *polA* PCR positive (range 5.4 – 38.920; mean 3.227 copies/ug total DNA). Among 8/14 samples that were fully typed, 7 strain subtypes (21a11/d, 129/f, 109/f, 1410f, 59/f, 149/f, 149/f) were identified with only 2 strains exhibiting identical patterns (21a11/d). The A2059G mutation was found in 1 specimen from an HIV-negative subject. Cali *Tp* strain types had a Shannon index of 2.2, higher than all 13 studies in other localities reported in two recent reviews.

**Conclusions** There is a high *T. pallidum* strain diversity in Cali, Colombia, probably due to population mobilisation and close proximity to other syphilis-endemic regions (i.e. Buenaventura, Pacific Coast). Contact tracing and cluster identification is difficult to achieve in this setting. Although our sample size was small, the identification of the A2058G mutation suggests that macrolides should be used with caution for syphilis treatment in Cali and close monitoring for macrolide resistant strains should be initiated.

**P3.274** CHARACTERIZATION OF VAGINAL LACTOBACILLUS AMONG NON-PREGNANT WOMEN WITH AND WITHOUT BACTERIAL VAGINOSIS IN INDIA AND US

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**Background** *Lactobacillus* is an integral part of vaginal microbiota that maintains healthy environment and plays an important role in preventing sexually transmitted infections and HIV. We profile the *Lactobacillus* species present when the women are healthy or have Bacterial Vaginosis among women in US and India.

**Method** Between February 2010 and November 2011, a sample of 75 women attending Prerana Women’s Health Clinic in India or San Francisco City Clinic in USA were sampled and diagnosed for BV based on Amsel’s Criteria. In addition, Gram stained smears of vaginal fluid were Nugent scored. Vaginal swabs were then cultured in MRS broth. Gram positive *Lactobacilli* generating about 600–800bp amplicon by16S rDNA PCR with 16S primers were further characterised by sequencing.

**Result** Vaginal samples were obtained from 75 women. According to Amsel criteria, 34 women were healthy and 41 women had BV. *Lactobacilli* were isolated from 22 healthy Indian and 10 healthy US women. *Lactobacilli* were also isolated from 4 Indian and 5 US women with BV. Eleven *Lactobacillus* species were isolated from 26 Indian women and 9 species were identified from 15 US women. The common *Lactobacilli* species found in Indian women included *L. crispatus* (24.3%), *L. gasseri* (24.3%), and *L. jensenii* (15.3%), while *L. crispatus* (52.0%), *L. jensenii* (20.0%), and *L. coleohominis* (12.0%) were common in US women. *L. crispatus* was cultured from 44% of healthy and 49% of women with BV. *L. jensenii*, *L. gasseri*, and *L. acidophilus* were cultured from 25.6%, 25.3% and 2.9% of healthy women; and 2.4%, 4.9% and 0.0% of BV women, respectively.

**Conclusion** Our findings showed *lactobacilli* species present in healthy vagina of women in India do not differ from those reported from other countries. This information is useful to development of microbicides for HIV prevention.

**P3.275** NEISSERIA GONORROEOEAE: SITUATION OF ANTI BiOTIC RESISTANCE IN GERMANY

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**Background** The dramatic development of antimicrobial resistance in *Neisseria gonorrhoeae* is a serious problem for treatment and control of gonorrhoea. Numerous formerly effective therapeutic agents are no longer appropriate.

High level penicillin resistance and quinolone resistance disseminated globally. The third generation cephalosporins are amongst the last agents to remain effective. Reduced susceptibility to these cephalosporins is increasingly common. Currently the emergence of *Neisseria gonorrhoeae* strains with reduced susceptibility to ceftriaxone can be observed. Azithromycin is an integral part of vaginal microbiota. Contact tracing and cluster identification is difficult to achieve in this setting. Although our sample size was small, the identification of the A2058G mutation suggests that macrolides should be used with caution for syphilis treatment in Cali and close monitoring for macrolide resistant strains should be initiated.