

all samples analysed. *T. pallidum* subtyping discriminated re-infection.

#### P1.45 EVALUATION OF SEKURE RPR REAGENT ON THE SK500 CLINICAL CHEMISTRY SYSTEM

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**Introduction** An automated and accurate laboratory assay would be of considerable utility to the diagnosis of syphilis and treatment follow-up. We compared the Sekure RPR (Rapid Plasma Reagin) test performed on the SK500 Clinical Chemistry System to RPR card test results.

**Methods** Serum samples were collected in the context of a 2 year observational cohort study of syphilis infected patients and controls. Syphilis was diagnosed using non-treponemal and treponemal testing. Sera collected at the time of diagnosis (M0) and at 3, 6, 9 and 12 months post-treatment were tested by a Macro-Vue RPR card test (RPR-C) (Becton Dickinson) and a Sekure RPR test (RPR-S) (Sekisui Diagnostics). RPR-S results are expressed in RPR units (R.U.), whereby 1 R.U. equals a 1-fold change in RPR-C titre. The agreement, linearity and reportable ranges were determined using RPR-C results as the gold standard. Linear regression was used to assess correlations from *before* and *after* implementation of an extra dilution step for samples with a strong suspicion of prozone effect.

**Results** In total, 451 samples from 150 participants were tested, including 120 new syphilis cases and 30 controls. All 30 controls tested negative. Initially there was a weak correlation between RPR-C and RPR-S values ( $r=0.15$ ). Further analyses identified 72 RPR-S samples with a strong suspicion of prozone effect. We therefore included an extra dilution step (10x) and retested 60/72 samples; values within the expected range were obtained for 58 of them. After implementing the extra dilution step the correlation was moderate ( $r=0.61$ ), increasing further to  $r=0.91$  for samples with RPR-C titres  $\leq 128$ . Of the 92 samples that tested RPR-C positive and RPR-S negative, 8 were from M0 (RPR-C: 1–4), which would have led to missed diagnoses.

**Conclusion** A reasonable correlation was found between the tested methods for mid-range RPR-C results (titre  $\leq 128$ ). However, prozone may occur in samples with high antibody concentrations. More investigation is required to elucidate the false negative RPR-S results.

#### P1.46 COMPARISON OF TWO ENZYME IMMUNOASSAYS FOR THE DETECTION OF IGG AND IGM ANTI-TREPONEMA PALLIDUM ANTIBODIES

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**Introduction** We aimed to compare two commercial enzyme immunoassays (EIA) for the detection of IgG and IgM anti-*Treponema pallidum* (Tp) antibodies.

**Methods** Serum samples were collected in the context of a larger study looking at diagnostic biomarkers for syphilis.

Patients with syphilis diagnosed by a treponemal and a non-treponemal assay, were followed for up to two years after treatment. Specimens collected at visit of diagnosis (B), and after three (M3) and six months (M6) of treatment were tested by EIAs detecting anti-Tp IgG and IgM from the manufacturers Euroimmun (EU) and Mikrogen (MI).

**Results** We tested 338 samples collected from 119 new syphilis cases (23 primary (P), 49 secondary (S), 31 early latent (EL), 16 late latent (LL)) and 30 uninfected controls. A total of 40 participants contributed to 1 sample, 29 to 2 samples and 80 to 3 samples. The controls contributed only to the samples collected at B. Overall 147, 86 and 105 samples were obtained at B, M3 and M6, respectively. The IgM assays were in agreement for 78,1% of samples; it varied according to the syphilis stage: P: 82,1%; S: 72,5%; EL: 80,8%; LL: 72,1% and decreased from B to M3: B: 84,4%; M3: 73,3%; M6: 73,3%. More samples tested positive with the MI (149) *versus* the EU (100) ( $p<0.001$ ). EU tested all control samples IgM negative, MI reported 1 positive and 1 borderline. The agreement of both IgG assays was 97,4%; it increased with the stage of infection: P: 91,1%; S: 97,7%; EL: 100%; LL: 100%, and over time: B: 95,2%; M3: 98,8%; M6: 99,0%. More samples tested positive with the EU (305) *versus* the MI (300) assay. EU reported all control samples IgG negative, MI detected 1 borderline sample.

**Conclusion** A good but not perfect agreement was observed for the EIAs detecting IgM. The agreement was highest in primary syphilis and lowest in late latent cases, and decreased over time of treatment. The MI IgM assay reported significantly more positive samples. Overall, we found a good agreement for the EIAs detecting IgG. Albeit that it was somewhat lower for primary syphilis and at baseline.

#### P1.47 DETECTION OF CERVICAL HUMAN PAPILLOMAVIRUS IN WOMEN ATTENDING FOR CERVICAL CANCER SCREENING BY VISUAL INSPECTION IN COTE D'IVOIRE

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**Introduction** Human Papillomaviruses (HPV) are small virus non-enveloped double-stranded circular DNA responsible of genital warts papilloma, precancerous lesions and cancers (cervix, vulva). In Côte d'Ivoire and many lower middle and incomes countries cervical cancer screening program based on visual inspection methods become the gold standard because cytology has shown many limits. This study aims to detect HPV DNA on women attending for cervical cancer screening program based on visual inspection by acid acetic and lugol (IVA/IVL).

**Methods** From March to December 2015, endocervical secretions from women attending cervical screening by IVA were submitted to HPV determination with PCR. HPV DNA was amplified using PGM09/11 primers which generated 450 base pairs at the L1 region. The samples harbouring HPV DNA were genotyped using the multiplex PCR with HPV 16, 18, 31, 33, 35, 45 and 51 primers.

**Results** The medium age of population was 32 years old. On 388 women enrolled in a visual inspection with acetic acid (VIA) program 5.8% were positif. HPV DNA was obtained in

9.02% of the population. A total of 31 (88.57%) specimens harbouring HPV DNA were genotypes using multiplex PCR versus 11.43%, which were not genotyped using HPV 16,18, 31, 33, 35, 45 and 51 by multiplex PCR. HPV genotyping gave 63 different HPV with 28.57% who had a single infection while 71.43% have a multiple infection. HPV genotypes prevalence were the followed: HPV 16 (28.57%), HPV 18 (23.80%), HPV35 (19.04%), HPV 45 (19.04%), HPV 51 (3.17%) and HPV 33 (1.58%). By using PCR as gold standard VIA sensibility was 16.12% and the specificity 95.45%.

**Conclusion** HPV circulate in Cote d'Ivoire in women attending for cervical cancer screening by visual inspection with acetic acid or lugol. Visual inspection with acetic acid or lugol seem to have a good specificity. HPV Genotypes 16 and 18 included in the vaccine available seem to be the most prevalent.

#### P1.48 ABSTRACT WITHDRAWN

#### P1.49 THE MUTATIONS ON GENES RELATED TO MACROLIDE OR FLUOROQUINOLONE RESISTANCE ON *M. GENITALIUM* IN JAPAN

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**Introduction** The condition of antimicrobial resistance in *Mycoplasma genitalium* has been becoming serious in the world. The macrolide-resistance is closely related to mutation on region of 23S rRNA gene. The fluoroquinolone-resistance is probably related to mutation on gyrase or Topoisomerase IV genes such as *gyrA* or *parC*, like as other fluoroquinolone-resistant bacteria. In our study, we analysed the mutations related to antimicrobial resistance among *M. genitalium* genes which collected in Japan and compared with mutations of M6489, the multidrug-resistant strain.

**Methods** The *M. genitalium* genomes were collected from the urine specimens of Japanese males with urethritis during the period between 2005 and 2016. In addition, the genomes of *M. genitalium* strain which can grow in the culture media, included M6489, the multidrug-resistant strain were used. The region V of 23S rRNA and quinolone-resistance determining region (QRDR) on *gyrA* and *parC* genes were sequenced and the mutations related to macrolide- or quinolone-resistance were analysed.

**Results** The *M. genitalium* genomes from 157 Japanese males and 10 *M. genitalium* strains were analysed. Among the genomes from Japanese males, mutations related to macrolide-resistance such as A2058G or A2059G were detected in 4.4% (4/90) genomes at 2005–2009 and in 40.3% (27/67) at 2010–2016. Two types of mutations on the *gyrA* gene with amino-acid change and 11 types of mutations on the *parC* gene with amino-acid change were found. These mutations were detected in 26.6% (24/90) at 2005–2009% and 53.7% (36/67) at 2010–2016. Most frequent mutations were Pro69→Ser in 18 genomes and Ser80→Ile in 16 genomes. M6489 had A2059G

on 23S rRNA and Asp87→Asn on *gyrA* and Ser80→Ile on *parC* gene. If these mutations on M6489 were related to fluoroquinolone-resistance, the fluoroquinolone-resistant *M. genitalium* increased 3.4% (3/87) to 16.4% (11/67).

**Conclusion** The mutations related to macrolide-resistance and fluoroquinolone-resistance genes increased in Japan.

#### P1.50 ABSTRACT WITHDRAWN

#### P1.51 ANTIMICROBIAL RESISTANCE (AMR) AND NG-MAST PROFILE OF *NEISSERIA GONORRHOEA* ISOLATES COLLECTED FROM STD PATIENTS FROM ACROSS INDIA

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**Introduction** *Neisseria gonorrhoeae* multiantigen sequence typing (NG-MAST) is a highly discriminatory technique for assessing the genetic diversity of *N. gonorrhoeae* (NG) and has also been put forward as a tool for predicting specific antimicrobial resistance (AMR) phenotypes. In light of the above, the present study was undertaken to investigate the molecular epidemiology of NG in India and to examine if it can be used as a means for predicting AMR.

**Methods** A total of 204 consecutive NG isolates collected between April 2010 - October 2013 were investigated. Antimicrobial susceptibility testing was done using disc diffusion method and E test and the results interpreted using the breakpoint criteria of CDS technique. NG-MAST was performed as described previously. WHO *N. gonorrhoeae* reference strains F, G, K-P were used as controls. Association between NG-MAST sequence type (ST) and antimicrobial susceptibility was probed using  $\chi^2$  and Fisher's exact tests.

**Results** Rates of resistance to classical antibiotics were high. Decreased susceptibility (DS) to ceftriaxone (MIC 0.032–0.25 µg/ml) was seen in 7.3% while azithromycin resistance (MIC ≥1 µg/ml) in 2.5% isolates. A total of 202 NG isolates were assigned into 108 different STs while 2 were not typable. The high genetic diversity arose from the allelic combination of 80 *por* and 44 *tbpB* alleles and the overall mean genetic distance was 85.5 (SE 4.6) nucleotide differences. Out of 108 STs, 84 (77.8%) were novel. The majority of STs (75.9%, 82 of 108) were represented by singletons, whereas the remaining STs included between 2 and 38 isolates. The most common STs were ST6058 (n=38, 18.8%), ST2990 (n=6, 2.9%), ST6069, ST9775, ST9783, ST9875 (n=5, 2.5% each). There was a significant association between ST6058 and resistance to penicillin (p=0.00) and tetracycline (p=0.00) and ST6069 and ST 6083 and DS to ceftriaxone (p=0.00 and p=0.01 respectively).

**Conclusion** The present study highlights a heterogeneous gonococcal population in India. Our data, although on a limited number of NG isolates, testify to an association between genotype and AMR phenotype.