

Nucleic Acid Amplification Tests (NAATs) for *Chlamydia trachomatis* (CT) and *Neisseria gonorrhoeae* (GC) have become routine. These methods are validated for use with urogenital samples and have a faster turn-around-time for results with automation. Testing of non-validated samples is common which raises concerns about assay performance. In Australia, the 2005 Public Health Laboratory Network (PHLN) guideline recommends repeat testing of all initial positive GC NAAT results with a suitable alternate NAAT assay. In this study, results from > 70,000 patient samples from 2 laboratories were reviewed in accordance with the PHLN recommendations. The laboratories used the APTIMA Combo 2 (AC2) and APTIMA *Neisseria gonorrhoeae* (AGC) assays with the PANTHER instrument.

72,253 AC2 results were available for analysis which included 1,174 (1.68%) initially AC2 reactive (positive or equivocal) samples which also had results for the AGC assay used as the confirmatory assay. For the reactive samples, the agreement between the AC2 and AGC assays occurred in 97.19% of samples; 1135 were positive in both assays and 6 were equivocal in both assays. Sample types tested included those with manufacturer's validation claims; urine, ThinPrep, vaginal, endocervical and urethral swabs and non-validated samples including throat, rectal, eye and joint fluids. Samples from throat swabs showed the highest numbers of discordant results followed by rectal swab samples. The percentage agreement of results obtained from all sample types was excellent, with an overall PPV of > 99%.

Without confirmatory testing, false positive results would have been reported for 13 samples representing 0.02% of all samples tested. This study demonstrates that the initial AC2 results can be accepted with high confidence.

#### P2.002 EVALUATION OF A REAL-TIME PCR-BASED TEST FOR BACTERIAL VAGINOSIS

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**Background** Bacterial vaginosis (BV) is the most common cause of vaginal discharge in women. Since BV is associated with significant morbidity, accurate tools for diagnosing the disease are important. The Amsel criteria (AC) and Nugent score (NS) are currently used for BV diagnosis. Recently, a number of PCR-based tests providing objective, sensitive and specific BV detection have been described. This study aimed to evaluate a newly developed BV test based on quantitative detection of *Gardnerella vaginalis*, *Atopobium vaginae*, *Lactobacillus* spp. and total Bacteria using multiplex PCR.

**Methods** PCR criteria were elaborated based on the relative counts of the targeted bacteria to classify vaginal microflora as BV, no BV or intermediate. Vaginal samples for the test evaluation were obtained from 274 patients addressing a gynaecologist for routine examination. All participants were of reproductive age, not pregnant and not menstruating at the time of enrollment. BV was diagnosed using the AC and NS.

**Results** According to the NS results, 66 patients were BV positive, 156 were BV negative, and 52 were classified as intermediate. All patients positive by the NS were positive by the AC, and all patients negative by the NS were negative by the AC. Among 66 BV positive samples, 62 demonstrated PCR results corresponding to BV, and 4 samples were BV negative by PCR (94% sensitivity). Of the 156 negative samples, 151 were interpreted as BV negative using PCR criteria, and 5 samples - as BV positive (97% specificity).

**Conclusion** A multiplex real-time PCR test for BV was developed showing 94% sensitivity and 97% specificity.

#### P2.003 FEASIBILITY AND ACCEPTABILITY OF SELF-COLLECTED VAGINAL SWABS FOR DIAGNOSIS OF BACTERIAL VAGINOSIS AMONG PREGNANT WOMEN IN A COMMUNITY SETTING IN RURAL MYSORE, INDIA

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**Background** Bacterial vaginosis (BV) is a common cause of adverse birth outcomes. Its association with other obstetric and gynecologic complications and HIV are increasingly recognised. This study examined the acceptability and feasibility of using self-collected vaginal swabs (SVS) for screening of BV in a community setting among rural pregnant women in Mysore Taluk, India.

**Methods** A community based cross-sectional study was carried out between June 2007 and December 2010. Mobile medical clinics offered antenatal care and HIV testing in all 144 rural villages in Mysore Taluk. Women were also screened for BV using SVS and asked about their experience with the collecting process. Gram-stain evaluation of vaginal samples using Nugent score (NS) was conducted by two trained laboratory personnel.

**Results** Among the 1675 women attending the mobile medical clinics from the 144 villages, 1541 (92%) were included in the analyses. 1639 agreed to provide vaginal swabs (97.8% response rate). The quality of the swabs was satisfactory in 1545 of the cases. There were 134 non-interpretable slides owing to poor quality. The median age of women was 20 years (range 14 to 40 years). Majority (98.7%) reported themselves as Hindus and 1634 (97.5%) were housewives. The prevalence of BV was 9.9% with a NS of 7–10 and 14.9% of women had intermediate flora on Nugent score of 4–6. While 212 women (12.9%) reported collecting the vaginal swab as being 'very easy', 1402 (85%) found it 'easy' and 22 (1.3%) reported it as 'difficult'. Only 12 women were unable to collect the swab for various reasons.

**Conclusion** These study results support the use of self-collected vaginal swabs for diagnosing BV. Self-collected swabs to detect BV were well accepted by most of rural pregnant women in this region, and the quality of the swabs seemed to be satisfactory.

#### P2.004 DOES BACTERIAL VAGINOSIS HAVE AN ASSOCIATION WITH SIL?

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**Background** Literature documents incomplete and conflicting evidence between bacterial vaginosis and its association with cervical intraepithelial neoplasia in cervicovaginal smears (CS).

**Aim** To investigate if there is an association between bacterial vaginosis (BV) and squamous intraepithelial lesion (SIL) in CS.

**Methods** A retrospective analysis of 6902 cervicovaginal cases accrued during January 2006 -December 2009 from an opportunistic screening programme was done. The cases diagnosed as unsatisfactory (n = 239), glandular cell abnormalities (n = 9) and carcinoma (n = 12) were excluded. Thus the study was based on 6642 cases. These were reviewed to identify SIL and BV. The correlation with histology was done wherever available. A diagnosis of BV was rendered if 10 clue cells per low power field and at least 5 fields showing cocco-bacteria were detected on Pap stained smears. Statistical evaluation was carried by applying Z test to determine if BV has an association to SIL in CS.

**Results** Of the 6642 cases analysed, BV was present in 13% cases and absent in 87% cases. SIL was reported in 7% BV present group and 2% in the BV absent group (P < 0.001). The difference between

the two proportions was highly significant. We also compared BV present in LSIL group with HSIL group. BV was present in 29% cases in LSIL group compared to 45% in HSIL group ( $P = 0.03$ ). The difference between the two proportions was significant.

**Conclusion** The presence of BV was significantly higher in SIL group and was associated with the severity of SIL. The presence of BV should not deter a diagnosis of SIL but trigger a meticulous search for abnormal cells.

**P2.005** **GIANT CONDYLOMA ACUMINATA OF BUSCHKE AND LOWENSTEIN TREATED SUCCESSFULLY SURGICALLY**

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**Introduction** The Buschke Lowenstein tumour is an extremely rare, slow-growing, locally destructive, cauliflower-like mass, also known as giant condyloma acuminata.

**Case Report:** We report a case of a 42 year old male who presented to the department of surgery with perineal tumour since two years. The mass was painless initially but later became painful. On local examination, the ano perineal region of the patient was completely occupied by a cauliflower like tumour with multiple fistulae. After histopathological confirmation, the tumour was removed surgically, as it was resistant to medical treatment. Radical surgery was attempted. The tumour was found to be very vascular and deeply infiltrating. Wide local resection of the perianal tissue was performed

**Discussion** Histopathology of BLT shows blunt-shaped masses of tumour project deeply into the dermis and contiguous structures. The tumour cells have little evidence of atypia and are not found inside blood vessels or lymphatics. Individual keratinocytes may show keratinization, but no horn pearls are seen. Lymphohistiocytic inflammation is usually present.

**Conclusion** Troublesome recurrences of BLT occur frequently and a propensity for infection and fistula formation is common. Regardless of the size and origin of BLTs, gaining early control of the disease using wide, radical surgical excision provides the best overall rate of survival.

**P2.006** **EVALUATION OF A MODIFIED CD4 COUNT METHOD FOR HIV MONITORING**

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**Introduction** The measurement of CD4 count for HIV-infected individuals relies on flow cytometry methods. The standard one is a single platform technology, which uses 50  $\mu$ l of samples and 10  $\mu$ l of monoclonal antibodies. To reduce the CD4 count cost, we propose to asses a modified method based on reducing the sample and monoclonal antibodies volumes.

**Methods** Between February and May 2011, we have tested 90 samples of HIV-infected persons by the standard method i.e using 50  $\mu$ l of anticoagulated blood and 10  $\mu$ l of monoclonal antibodies (CD3FITC/CD4PE/CD45PerCP, Becton-Dickinson) and by the modified method i.e using 20  $\mu$ l of anticoagulated blood and 2  $\mu$ l of monoclonal antibodies (CD3FITC/CD4PE/CD45PerCP, Becton-Dickinson), in our laboratory. The % of CD4 as well as the absolute count (TrueCount, Becton-Dickinson) was determined for both methods by using Cellquest-Pro on FacsCalibur (Becton-Dickinson). Linear regression and Bland and Altman analysis were performed to assess correlation and agreement between both methods.

**Results** When analysing the whole sample, the modified method showed a strong correlation with the standard method,  $r = 0.99$  for CD4 count percent. Bland and Altman analysis revealed a mean bias

of  $-0.1\%$  (Limit of agreement:  $-3.0, 2.8$ ). Regarding the absolute count of CD4,  $r$  was 0.99 and the mean bias was 9 cells/ $\mu$ l (LOA:  $-64.8, 82.5$ ). When the statistical analysis is performed for the strata of  $CD4 \leq 350$ , the  $r$  was 0.98, and the mean bias was  $-1$  cell/ $\mu$ l (LOA:  $-44.1, 42.2$ ).

**Conclusion** The modified method based on reducing blood and antibodies volumes showed similar results to the standard method. This low cost method may be an interesting alternative method to measure CD4 count in developing countries.

**P2.007** **GENETIC RISK OF DNA REPAIR GENE POLYMORPHISMS IN HIGH-RISK HPV ASSOCIATED CERVICAL CARCINOGENESIS**

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**Background** Cervical cancer is the second most common cancer in women worldwide. A large number of young sexually active women get infected by human papillomavirus (HPV) but only a small fraction of them have persistent infection and develop cervical cancer pointing to co-factors including host genetics that might play a role in outcome of the HPV infection. This study was designed to examine the polymorphisms associated with four DNA repair genes, viz., XRCC1 (Arg194Trp, Arg399Gln and Arg280His), ERCC1 Asp118Asp, ERCC2 Lys751Gln and ERCC4 Arg415Gln and investigate their role as susceptibility markers for cervical precancer (LSIL & HSIL) and cancer.

**Methods** The cases comprised 105 patients: 65 cervical squamous cell carcinomas (SCCs), and 40 squamous intraepithelial lesions (SILs). 65 healthy women were recruited as the controls. Genotypes were determined by PCR-RFLP and DNA sequencing techniques.

**Results** Our data showed a positive association between the polymorphisms of codons 194 ( $p = 0.001$ , OR = 22.4, 95% CI = 9.15–55.03), 280 ( $p = 0.001$ , OR = 20.04, 95% CI = 8.4–47.5) and 399 ( $p = 0.001$ , OR = 11.11, 95% CI = 4.98–24.78) and cervical cancer. SIL patients also showed a significant association with codon 194 ( $p = 0.001$ , OR = 7.56, 95% CI = 3.42–16.70) and 280 ( $p = 0.015$ , OR = 3.05, 95% CI = 1.35–6.88) but not with 399 ( $p = 0.142$ ). Positive correlation was also found in ERCC4 Gln415Gln in both SCCs and SILs ( $p = 0.001$ , OR = 5.45, 95% = 3.19–9.29 and  $p = 0.001$ , OR = 2.76, 95% = 1.55–4.91, respectively). For ERCC2 Gln751Gln the association was significant for SCCs ( $p = 0.010$ , OR = 1.44, 95% = 0.86–2.14) but not for SILs ( $p = 0.088$ ). However the risk for cervical precancer and cancer did not appear to differ significantly amongst individuals featuring the ERCC1 Asp118Asp genotype ( $p = 0.594$  and  $0.080$ , respectively).

**Conclusion** We analysed the association between XRCC1, ERCC4, ERCC2 and ERCC1 polymorphisms and the individual susceptibility to develop cervical precancer and cancer. We attempt to contribute to the discovery of which biomarkers of DNA repair are useful for screening this high-risk population for primary preventing and early detection of cervical cancer.

**P2.008** **MISTAKEN CASE OF CHILD ABUSE: A CASE REPORT**

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**Introduction** The diagnosis of child abuse is based on a combination of child's history, physical findings, and when appropriate, laboratory and other tests. Overall, the diagnosis is often complicated but suspicion should always be followed by further investigations. Formulating a conclusion and reaching a diagnosis of child