ROLE OF TYPE SPECIFIC HERPES SIMPLEX VIRUS SEROLOGY IN DIAGNOSIS OF PRIMARY AND RECURRENT GENITAL HERPES: A STUDY IN THE INDIAN POPULATION

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Background Type specific serologic tests for herpes simplex virus (HSV)-1&2 are used for sero-epidemiological studies. Its early application has also been shown to be of benefit in testing for primary & recurrent infections in diagnosis and patient management. The present study was conducted to evaluate the role of type specific HSV-1&2 antibody detection as a diagnostic modality in patients with clinically suspected genital herpes.

Methods Sera obtained from 44 patients with clinically suspected genital herpes attending the STD clinic of our institution over last 7 months were tested for type specific HSV-1&2 IgG antibody by glycoprotein G based ELISA (HerpeSelect 1&2, Focus Diagnostics, USA). Direct Fluorescent Antibody testing (DFA) (BIO-RAD laboratories, USA) was used for identification and typing of HSV-1&2 in genital ulcer specimens in all cases.

Results There were 21 cases of primary & 23 cases of recurrent genital ulceration. DFA was positive in 68% samples (43% only HSV-2, 9% only HSV-1, 16% both HSV-1&2). On comparison with DFA, the sensitivity, specificity, positive and negative predictive value of HSV-2 IgG ELISA in primary herpetic ulcer was 8%, 75%, 33%, 33%, while it was 79%, 89%, 92%, 73% in recurrent genital ulceration. The sensitivity, specificity, positive predictive and negative predictive values of HSV-1 IgG ELISA in primary herpetic ulcer was 50%, 67%, 37.5%, 77%, while it was 80%, 39%, 27%, and 87.5% in recurrent genital ulceration.

Conclusion Our results highlight the importance of HSV-2 IgG detection in strengthening the diagnosis of recurrent HSV-2 disease, while absence of HSV-2 IgG antibody helps in excluding genital herpes as a likely cause of recurrent genital ulceration. The identification of HSV-1 IgG antibody may not be useful for diagnosis in patients of genital ulcer disease highlighting the need for typing HSV-1 strains from the genital lesion.

COPAN ESWAB™, A LIQUID BASED MICROBIOLOGY DEVICE, CAN BE USED FOR THE PRESERVATION OF NEISSERIA GONORRHOEAE FOR CULTURE AND FOR DETECTION OF CT/GC BY GEN-PROBE® APTIMA® COMBO 2 ASSAY


Background Copan Liquid Amies Elution Swab (ESwab™) Collection and Transport System incorporates maintenance medium which can sustain the viability of Neisseria gonorrhoeae (NG). Swab specimens placed in the tube and stored at room temperature (RT*) can be cultured within 24 hours, and aliquots for nucleic acid amplification tests (NAATs) can be processed within five days.

Methods Clinicians at a Baltimore City Health Department STD clinic used standard-of-care aluminium shaft swabs with polyester tips to collect urethral swabs from men, smeared a slide for Gram stain, and placed NG positive (by smear) swabs into investigational ESwab™ tubes. The ESwab™ collection swab applicator was then broken off into the tube and the cap was closed. Culture was performed after tube sat from 5 to 28 hours at RT*. Tube was vortexed for 5 seconds; 100 μl was inoculated to MTM plate, streaked for isolation, and incubated in appropriate conditions for 48–72 hours; 100 μl was placed into Gen-Probe® Unisex transport tube for testing by APTIMA® COMBO 2 assay.

Results Of 35 swabs, 26 (74%) were culture positive for presumptive NG (Gram negative diplococci, oxidase positive). Colony counts ranged from 1 to > 500 colonies.

35 (100%) were positive for NG by Gen-Probe® APTIMA® COMBO 2 assay. Six (17%) were also positive for Chlamydia trachomatis (CT) by Gen-Probe® assay.

Conclusion Although quality of collected clinical specimen and quantity of NG on the swab are significant variables in obtaining reliable culture results, the ESwab™ System was able to maintain NG viability for at least 24 hours, and stabilised nucleic acids for Gen-Probe® testing. The ability to maintain culture viability of NG up to 24 hours until NAAT testing is completed could eliminate the necessity to perform cultures on all patients who are being screened for NG, with cultures maintained only for susceptibility testing for NAAT+ patients.

DIAGNOSIS OF PHARYNGEAL AND RECTAL NEISSERIA GONORRHOEAE INFECTIONS

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Background Despite nucleic acid amplification tests (NAAT) are widely used to detect Neisseria gonorrhoeae infections, so far no commercial kit has been cleared for testing rectal or pharyngeal swab samples, even if anal and/or oral sex practices are common.

In this study, a comparison between Real Time PCR Versant CT/ GC DNA 1.0 (Siemens) and N. gonorrhoeae culture performances has been conducted, testing rectal or pharyngeal secretions collected by E-swabs (Cowan).

Methods Study group. A prospective study was performed with 171 subjects (130 males and 41 females) attending the STD Outpatients Clinic of St. Orsola Hospital, Bologna. All the patients were enrolled because having unsafe receptive anal and/or pharyngeal sex intercourse.

NAAT methods. All the specimens were tested by Versant CT/ GC DNA 1.0. As a confirmation, all the specimens scored positive for N. gonorrhoeae were retested, using the same extraction, by a "home-made" PCR assay, targeting cppB gene.

N. gonorrhoeae culture. Bacteria were isolated in Thayer-Martin medium and identified by API NH assay (bioMérieux). Antimicrobial susceptibility was assessed by Kirby-Bauer Test.

Results A total of 227 samples were obtained. In particular, 56 patients provided both the specimens, 89 patients provided only pharyngeal swabs, whereas only rectal specimens were collected from the remaining 26 patients.

Versant CT/GC DNA 1.0 gave positive results for N. gonorrhoeae in 13 pharyngeal in 7 rectal samples, all from MSM. All the Versant reactive results were confirmed by “home-made” PCR. Prevalence of rectal infection was 8.5% (7 positive out of 82 patients), whereas prevalence of pharyngeal infection was 9.0% (13/145). Culture was far less sensitive than NAAT, since only 4 samples were identified. All of them were resistant to quinolones, but susceptible to cephalosporins (ceftriaxone and cefixime).

Conclusions Pharyngeal and/or rectal screening for gonorrhoea should be considered essential in consultations for MSM in STD clinics.

EVALUATION OF ELECSYS® IMMUNOASSAY SYSTEM FOR DETERMINATION OF TYPE-SPECIFIC IgG ANTIBODIES TO HSV-1 AND HSV-2


P5.068

P5.069

P5.070

P5.071
Background HSV infections during pregnancy are generally asymptomatic and associated with high rate of neonatal morbidity and death. Differential serologic diagnosis of HSV infections in pregnancy is important for the correct assessment of the infection status. The aim of this study was to evaluate the performance of Roche Elecsys® assays for the type specific determination of HSV-1 and HSV-2 IgG antibodies.

Methods A total of 800 samples obtained from sexually active adults (n = 300), pregnant women (n = 400) and herpes infection suspected patients (n = 100) were analysed using Roche Elecsys® HSV-1 and HSV-2 assays and commercially available Focus Diagnostics HerpeSelect, Radim HERPES S.V and Diasorin LIAISON type specific ELISA assays.

A commercially available Western Blot assay was used for the resolution of the discrepant results.

For further confirmation of the type specific detection performance of the Elecsys® HSV-1 and HSV-2 assays 310 potentially cross reactive samples for HSV-2 and 123 samples for HSV-1 were analysed.

Results The Elecsys® HSV-1 IgG assay showed a relative sensitivity of 95.6 to 100%. The relative specificity was between 97.6 and 100%. The Elecsys® HSV-2 IgG assay showed a relative sensitivity of 92.6 to 100%. The relative specificity was between 98.7 and 100%. These assays exhibit excellent differentiation between HSV-1 and HSV-2 infections with no cross reactivity to other herpes viruses like CMV, VZV or EBV.

Conclusion The results obtained with the type specific Roche Elecsys® HSV-1 and HSV-2 assays indicate that these assays are useful, specific and sensitive for the differential determination of HSV-1 and HSV-2 IgG antibodies in serum or plasma samples. The advantage of the Roche Elecsys® assays compared to ELISA based assays, is that these assays are rapid and can be performed in a fully automated process.

P5.072 THE PERFORMANCE OF TWO IGG ELISA METHODS TO DETECT HSV-2 INFECTIONS AMONG SOUTH-AFRICAN WOMEN WHO ARE AT HIGHER RISK OF BECOMING HIV INFECTED


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Background Specimens collected in Pretoria for the FEM-PrEP study, a phase III trial on pre-exposure prophylaxis for HIV prevention among African women, were tested for Herpes Simplex Virus type II (HSV-2). We present here the performance of the Focus HerpeSelect and Kalon HSV-2 gG2 ELISA.

Methods The HSV-2 infection was determined in 701 women at baseline with two IgG ELISAs: Kalon HSV-2 gG2 ELISA (Kalon Biologicals Ltd.) and HerpeSelect HSV IgG ELISA (Focus Technologies). Participants were considered true positive for HSV-2 when specimens were reactive in both assays. In order to determine incident HSV-2 infections during the study, specimens collected at final visit (i.e. after 52 weeks/at product interruption visit) of participants being HSV-2 seronegative at baseline were tested using the same assays.

Results At baseline, 287 and 315 positive results were found using the Kalon- and Focus assay, respectively. All Kalon positives were also positive in the Focus assay and considered to be true infections (initial prevalence: 40.9%). Of the 28 specimens positive with the Focus only, 10 of them became true positive at final visit. We therefore assume that the Kalon missed 10 infections and, the Focus detected falsely 18 positives at baseline, resulting in a final HSV-2 prevalence of 42.4% at baseline. At final visit, an additional 33 new infections were found. At baseline we obtained a sensitivity of 100% (95% CI: 98.8–100) and 96.6% (95% CI: 93.9–98.4) and a specificity of 95.5% (95% CI: 93.1–97.5) and 100% (95% CI: 99.1–100) for Focus and Kalon respectively.

Conclusion Although our study confirms the assay performance findings of previous studies in Sub-Saharan countries, we found less pronounced differences in terms of sensitivity and specificity of both assays using the cut-off as prescribed by the manufacturers. The prevalence of HSV-2 found in our study corresponds to previously reported results.

P5.073 COMPARISON OF COBAS® 4800 HPV ASSAY TO DIGENE HYBRID CAPTURE 2, ROCHE LINEAR ARRAY, AND AMPICOR IN THE DETECTION OF HIGH-RISK HUMAN PAPILLOMAVIRUS GENOTYPES IN WOMEN WITH PREVIOUS ABNORMAL PAP SMEARS


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Introduction Cobas® 4800 HPV assay has been evaluated as a screening and triage application recently. The aim of this study was to evaluate the performance of the Cobas® 4800 HPV assay for the detection and identification of high-risk (HR) HPV genotypes after treatment of high grade lesion by comparison with the Hybrid Capture 2® (HC2), Amplicor (Amp), and Linear Array (LA) HPV tests.

Methods Four hundred and six PreservCyt® specimens from women undergoing management for a high-grade Pap abnormality were evaluated and results compared with the HC2, Amp, and LA HPV test results, in addition to histological diagnosis of a contemporaneously collected biopsy.

Results The sensitivity in detection of underlying high-grade histological diagnosis by Cobas® 4800 HPV was 90.6%, for HC2 86.1%, whilst for Amp and LA 92.9% and 91.8% respectively. Restricting detection of Cobas® 4800 HPV to only types 16 and 18 resulted in sensitivity of 60.0%. Detection of HR genotypes by Cobas® 4800 HPV showed a concordance of 86.9%, 96.1%, and 96.3% when compared to HC2, Amp and LA respectively. Detection of HPV 16 and 18 by Cobas® 4800 HPV showed a concordance of 97.3% and 99% respectively when compared to LA.

Conclusion The performance of Cobas® 4800 HPV was equivalent to the Amp and LA HPV tests for HR HPV detection. Cobas® 4800 HPV identified more underlying histologically-confirmed high-grade lesions than the HC2 HPV test, with the added advantage of identifying HPV 16 and 18 genotypes present.

P5.074 IMMUNE ACTIVATION AFTER STIMULATION WITH CRYPTOCOCCUS NEOFORMANS ANTIGENS PRE AND POST ART INITIATION IN HIV-1 POSITIVE UGANDANS


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Background Cryptococcus meningitis immune reconstitution inflammatory syndrome (CM-IRIS) is a medical condition that complicates recovery from immunodeficiency as a result of anti retroviral therapy (ART) in patients living with Human Immunodeficiency Virus type 1 (HIV-1) in the sub Sahara Africa region.