

P2.094 THE CONTRIBUTION OF MACROLIDE RESISTANCE MUTATIONS TO FAILURE OF AZITHROMYCIN TREATMENT IN MYCOPLASMA GENITALIUM INFECTION

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Background Current recommended treatment for *Mycoplasma genitalium* (Mg) is azithromycin. Macrolide resistance mutations (MRM), predominantly on the 23S rRNA gene of Mg, have been found to be associated with failure of azithromycin. We aimed to determine the efficacy of 1g-azithromycin in a prospective cohort of Mg-infected STI clinic attendees, and to determine the contribution of MRM to treatment failure.

Method We commenced an observational study in July 2012 in which symptomatic patients diagnosed with Mg by PCR at Melbourne Sexual Health Centre are retested for Mg 14 and 28 days following treatment with 1g-azithromycin. Testing for MRM using high-resolution melt analysis (HRM) is conducted on day 0 and on positive samples at days 14 and 28. Participants are managed on the basis of clinical symptoms and not detection of MRM. Study will complete, May 2013.

Results 105 participants have been recruited; 89 have completed all study requirements. 48/89 (54%; 95% CIs 44–64%) participants were Mg PCR negative at day 28. 41/89 (46%; 95% CIs 36–56%) did not respond to 1g azithromycin: 11/41 (27%) had a persistently positive Mg PCR on day 28 without reported risk of re-exposure (*presumptive failures*) and 30 of 41 (73%) had persistent symptoms of MG prior to day 28 and required interim treatment with moxifloxacin (*probable failures*). Of the 41 failures, 40 (98%) had MRM detected: 30 (75%) at baseline and 10 (25%) at day 14 only. Of the 48 azithromycin-responders 4(8%) had MRM detected at baseline.

Conclusion The azithromycin cure rate for Mg in this clinic cohort was only 54%. MRM were detected in virtually all cases of azithromycin-failure, and were uncommon in azithromycin-responders. The majority of MRM were detected prior to treatment. These findings have implications for the use of macrolides as current recommended treatment for *M. genitalium*, and highlight the need for evaluation of alternative treatment approaches.

P2.095 ORIGINS OF REPEAT INFECTIONS WITH MYCOPLASMA GENITALIUM (MG) AMONG HETEROSEXUAL MEN IN TWO SOUTHERN U.S. CITIES

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Background The purpose of this study was to examine the origins of repeat infections with Mg among men. High repeat infection rates have been consistently reported and treatment failure secondary to macrolide resistance is thought to be the primary cause. This study adds to the growing literature by describing the possible origins of repeat Mg infections in among men, primarily African American, attending two public STD clinics in southern cities in the U.S.

Methods Men diagnosed with NGU at an STD clinic in New Orleans, Louisiana, and Jackson, Mississippi, were tested for Mg using the GenProbe research-use-only assay Mg+ men underwent a 4–10 week test of cure visit (TOC) following treatment with 1 g

azithromycin. Detailed sexual behaviour data were collected at baseline and follow-up via ACASI and genotyping was performed.

Results Of 205 men with MG, 135 returned for TOC visit and of those, 34.3% were positive. Of the 46 who were positive at TOC, 19.6% reported sexual re-exposure to a baseline partner, 6.5% reported sexual exposure to a new partner, 6.5% reported sexual exposure to both, and 67.4% denied sexual re-exposure. Men who re-tested positive for Mg at TOC (n = 46) were no more likely than those who tested negative (n = 88) to have had sexual exposure to a baseline partner (31.8% vs. 26.1%, P = 0.55) or exposure to a new partner (21.4% vs. 11.3%, P = 0.22). Genotyping on baseline/TOC positive pairs is being performed and will be used in conjunction with behavioural data to more precisely estimate the treatment failure rate.

Conclusion The TOC repeat infection rate among men with Mg is high. Our data are consistent with the published literature corroborating that repeat infections in men treated with azithromycin is usually due to treatment failure rather than re-infection. Research is needed to optimise treatment of Mg infections in men.

P2.096 EVALUATION OF GONOCOCCAL RESISTANCE IN AUSTRIA

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Background The Austrian Society for Sexually Transmitted Diseases started a surveillance project in 2010 to evaluate the gonococcal resistance in Austria.

Methods In 2010 and 2011 a national network of 39 centres was established to collect 1569 gonococcal isolates and the anamnestic data of the patients. Gonococcal culture plates were sent from the participating laboratories to the Outpatient's Center in Vienna, where gonococcal resistance testing was performed by disc diffusion test, agardilution breakpoint technique, agardilution, and Etest. The following antibiotics were tested: cefixime, ceftriaxone, penicillin, ciprofloxacin, azithromycin, tetracycline, spectinomycin, and gentamicin. Results were interpreted according to CLSI and EUCAST guidelines.

Results In Vienna, 1456 isolates were collected, whereas 111 strains were sent from the federal states. Of all collected isolates 87% were genital, 5% pharyngeal, and 7% rectal isolates, respectively. Gonococci were collected more often from men (56%) than from women, 10% of men reported homosexual contacts. A concurrent infection with *Chlamydia trachomatis* was observed in 15% of all patients.

While 2010 all isolates displayed susceptibility to third generation cephalosporines, in 2011 7 gonococcal strains were resistant to cefixime (MIC > 0.125 µg/ml) but still susceptible to ceftriaxone (MIC ≤ 0.125 µg/ml). Furthermore, an increase of MIC values for cefixime as well as for ceftriaxone was observed in 2011. Resistance to azithromycin increased from 1% in 2010 to 1.5% in 2011. Resistance to quinolones was detected in 58% in 2010 rising to more than 60% in 2011, respectively.

Conclusion Third generation cephalosporines still represent the most appropriate drug for gonococcal therapy. As Azithromycin resistance is low in Austria it is suitable for alternative therapy especially in case of coinfection with *C. trachomatis*. Due to the increasing MIC values for cephalosporines and the rising rates for

azithromycin resistance it is highly recommended to determine the resistance pattern of the respective gonococcal strain by culture performance.

P2.097 PCR FOR DIRECT DETECTION OF THE MOSAIC *NEISSERIA GONORRHOAE* *PEN*A GENE IN URINES AND CERVICAL, RECTAL AND TONSILLAR SWABS

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Introduction The mosaic *penA* gene, partly derived from commensal *Neisseria* strains, is strongly associated with diminished susceptibility of *Neisseria gonorrhoeae* (Ng) against cephalosporins. We developed a direct PCR test for Ng-positive clinical specimens to detect the mosaic *penA* gene.

Methods Swabs and urines from patients with gonorrhoea were in medium for NAAT testing (Aptima Combo 2). Corresponding Ng strains were obtained by culture on selective GC agar plates and stored at -80°C. Presence of a mosaic *penA* gene in these strains was demonstrated by PCR.

Results Using one conserved forward primer and two reverse primers, specific for mosaic- and wild type *PenA* genes, and SYBR green as a fluorescing agent, two real-time PCRs were developed. Testing diluted DNA samples showed that the mosaic *penA* gene PCR was 10–100 fold more sensitive than the wild type gene PCR. Both PCRs were negative with strains belonging to *N.meningitidis* (n = 3), *N.lactamica* (n = 4), *N.subflava* (n = 2), *N.cinerea* (n = 1) and *N.elongata* (n = 1). Ten urine (U), 10 cervical (C), 10 rectal (R) and 10 tonsillar (T) samples, all negative in the NAAT for Ng, were negative in both PCRs. Testing paired samples from patients, who had a positive culture and NAAT (10 R, 9 U, 8 C, 9 T) showed concordant results in 35/36 samples: 4 pairs tested positive in the mosaic PCR and 31 in the wild type PCR. From one patient a wild type strain had been cultured from the throat, but both *PenA* PCRs on the swab were negative, possibly due to a low amount of DNA.

Conclusion We successfully developed discriminating PCRs with which the Ng mosaic *penA* gene can be detected without culture of Ng. This test can be used to estimate the prevalence of diminished susceptibility of Ng against cephalosporins in regions where culture is no longer performed.

P2.098 SPECIATION AND ANTIFUNGAL SUSCEPTIBILITY TESTING OF CANDIDA SPECIES CAUSING ORAL THRUSH IN HIV PATIENTS

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Purpose Oral thrush by *Candida* species is a common ailment of individuals suffering from HIV infection. These species show a high resistance to antifungal drugs used for the treatment. Hence our study was conducted to determine the aetiology and antifungal susceptibility patterns of *Candida* isolates causing oral thrush in HIV patients.

Materials and Methods: Isolation of *Candida* species was attempted from 60 cases of oral thrush in HIV infected patients at the Department of Microbiology, Kasturba Medical College, Mangalore. Isolates were identified to species level based on chlamydo-spore formation; ability to form germ tube; assimilation/fermentation of carbohydrates; production of urease enzyme; formation of pellicle/surface film on Sabouraud's dextrose broth;

growth on Sabouraud's Dextrose Agar (SDA) with cycloheximide and growth on SDA at 37°C and 45°C. Antifungal drug susceptibility testing was done by macro broth dilution test using azole group such as fluconazole, itraconazole and ketoconazole.

Results 56 *Candida* species were isolated of which *C.albicans* was the predominant isolate (84%), followed by *C.tropicalis* (8%), *C.glabrata* (3.5%), *C.parapsilosis* and *C.kefir* (1.8% each). Most isolates (53) showed significantly higher resistance to fluconazole than the standard pathogenic control strain *C.albicans* NCPF 3153A. 31 isolates (66%) of *C.albicans* had Minimum Inhibitory Concentration (MIC) values 8 times that of control for ketoconazole. 23 isolates had MIC for itraconazole of 0.5 µg/ml which was only twice as high as that of control (0.125 µg/ml), all others having comparatively equivalent MIC to itraconazole.

Conclusion Our study indicates that although *C.albicans* is the predominant species, there are other species prevalent and causing infection in our HIV infected population. MIC's of our *Candida* isolates to commonly used antifungals such as fluconazole, ketoconazole and itraconazole were significantly higher than the control strain used in the study. Our study indicated that itraconazole was the most effective among the azole group of drugs.

P2.099 VEHICLE ALTERATIONS IN PODOPHYLOTOXINE TREATMENT: A PARTIAL DISAPPOINTMENT

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Background Podophylotoxine o, o5% gel is a routinely used for condylolma accuminatum treatment as home used procedure. However, irritations, sometimes severe are common. This may be due partially to leakage of the preparation to the surrounding non-infected tissue. It is difficult to expect the preparation to remain dry and only on affected areas in non-circumcised males and females.

Methods :We used compounded podophylotoxin o, 15% in adhesive creamy base. The team work with pharmaceutical technologist helped to create an adhesive creamy paste, with greater stability and uniformly distribution on individual lesions. The melting of the preparations was minimised by tailored compounding.

Results The vast majority of patients preferred cream to gel, both to far less irritations and excellent tolerability. Unfortunately, the overall success with cream formulation was disappointing. Therapeutic results seemed to be better, at the very beginning of the treatment, dye to the constant and prolonged delivery on the treated lesions. However, recurrences are far more frequent and tend to develop earlier than with gel podophylotoxin formulation.

Conclusion Probably, the cream formulation does have a therapeutic advantage in perianal region, because of the better adherence of the vehicle, and, when in out of office settings, of less irritation to surrounding tissue in the presence of over-applying the medicine, which frequently is the case.

P2.100 CLINICAL EFFICACY OF SITAFLOXACIN 100MG TWICE DAILY FOR 7 DAYS FOR PATIENTS WITH NON-GONOCOCCAL URETHRITIS

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To date, the standard treatment for the patients with chlamydial non-gonococcal urethritis (NGU) remains effective; however, conventional quinolone antibiotics have less activity against *Mycoplasma*