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Co-occurrence of Urogenital Mycoplasmas and Group B Streptococci with Chlamydial Cervicitis

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A b s t r a c t

The aim of our study was to evaluate whether in women with chlamydial cervicitis urogenital mycoplasmas and group B streptococci (GBS) colonization is found more often than among women with non-chlamydial cervicitis. This study included 351 (mean age 31.7 ± 6.82) not pregnant, menstruating, sexually active women. We confirmed a high frequency (49.3%) of *C. trachomatis* infection among women with cervicitis. Cervical ectopia was confirmed in 26.5% of examined women, in half of them ectopia was associated with chlamydial infection. We did not notice differences in frequency of colonization by urogenital mycoplasmas and GBS among women with chlamydial and non-chlamydial cervicitis.

K e y w o r d s: Chlamydia trachomatis, GBS, urogenital mycoplasmas, cervicitis, ectopia

Urogenital mycoplasmas and group B streptococci (GBS) are microorganisms colonizing female urogenital tract and playing an important role in the pathology of fetus and newborn. Urogenital mycoplasmas are often isolated, even in 54% of tested sexually active women of childbearing age (Schlicht *et al.*, 2004).

Sexual transmission of GBS is questioned (Honig *et al.*, 2002), while it is recognized in urogenital mycoplasmal infection (Keane *et al.*, 2000, Nunez-Troconis, 1999).

Today *Chlamydia trachomatis* is on the first place among sexually-transmitted bacteria (Millman *et al.*, 2004). Cervicitis, often with co-occurring ectopia, is the dominating clinical finding during *C. trachomatis* infection (Critchlow *et al.*, 1995, Giedrys-Kalemba *et al.*, 1994). Thus, the aim of our work was to evaluate, whether in women with chlamydial cervicitis urogenital mycoplasmas and GBS colonization is found more often than among women with non-chlamydial cervicitis.

This study included 351 (mean age 31.7 ± 6.82) not pregnant, menstruating, sexually active women who attended the Department and Clinic of Gynecology and Endocrinology, Medical University of Silesia in Katowice between 2001 and 2004. Cytological examination of cervix was performed in each case. All studied women had symptoms of cervicitis: mucopurulent endocervical discharge and/or greater or equal to 30 neutrophils per × 1000 field on the cervical Gram stain, and/or bleeding contact.

Patients with gonococcal infection and those receiving antibiotic therapy within the month before consultation were excluded from the study.

Sterile swabs were used to obtain material for testing/culturing of expected microorganisms (Friedek *et al.*, 2004). First swab (no 1) from vaginal fornix for GBS culturing was inoculated on Columbia sheep blood agar plate and incubated aerobically for 24–48 hours at 37°C. Identification of GBS was based on latex Slidex Streptokit (bioMerieux, France). Susceptibility of isolated GBS to antibiotics (ampicillin,

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erythromycin and clindamycin) was examined by disk-diffusion method. Second swab (no 2) from endocervical canal for isolation of genital mycoplasmas was inoculated in urea-arginine-broth transport medium (bioMerieux, France). Culturing of genital mycoplasmas was performed by using Mycoplasma IST (bioMerieux, France), according to manufacturer's instruction. Mycoplasma IST shows good sensitivity and specificity for *U. urealyticum* (100% and 90%, respectively), and for *M. hominis* (100% and 85% respectively) (Rastawicki *et al.*, 2004). Third swab (no 3) from endocervical canal for detection of *C. trachomatis* was fixed by acetone on a slide. Chlamydia Direct IF – DIF (bioMérieux, France) was used for *C. trachomatis* antigen detection, according to manufacturers instruction. Slides were examined in Nikon Model HB – 10101AF fluorescent microscope (x40 objective).

In studied group chlamydial etiology of cervicitis was confirmed in 49.3% (173/351). Genital mycoplasmas were isolated in 25.9% of women with cervicitis. There were statistically insignificant differences between occurrence of urogenital mycoplasmas in women with and without chlamydial cervical infection. *U. urealyticum* was a more frequently isolated species than *M. hominis* (Table I).

Table I
Prevalence of cervical ectopia, urogenital mycoplasmas and GBS among women with chlamydial and non-chlamydial cervicitis

	C. trachomatis – positive women (n = 173)		*C. trachomatis* – negative women (n = 178)	
	No	%	No	%
ectopia	42	24.3	51	28.7
Ureaplasma urealyticum	36	20.8	31	17.4
Mycoplasma hominis	4	2.3	3	1.7
Ureaplasma urealyticum and Mycoplasma hominis	7	4.0	10	5.6
GBS	23	13.3	26	14.6

The frequency of GBS isolation was 13.3% in chlamydia-positive and 14.6% in chlamydia-negative women. All isolated GBS strains were sensitive to ampicillin, only 9.6% of strains were resistant to erythromycin and 7.7% – to clindamycin.

Cervical ectopia was confirmed in 26.5% (93/351) of examined women. In 42 out of them ectopia was associated with cervical chlamydial infection.

In regions, where early screening program for detection of *C. trachomatis* infection was established, percent of infection is very low: in the USA – 4.7%, in Sweden – 5.4%, in Norway – 2.4% (Bakken *et al.*, 2004, Egger *et al.*, 1998, Miller *et al.*, 2004). In Poland, frequency of *C. trachomatis* infection in studied groups of symptomatic and asymptomatic women is around 20–40% (Choroszy-Król *et al.*, 1994, Giedrys-Kalemba *et al.*, 1994, Zbroch *et al.*, 2004). It is a well-known fact that cervicitis may be a predisposing factor for cervical ectopia (Critchlow *et al*., 1995, Giedrys-Kalemba *et al.*, 1994). In our study in 45.2% of women with cervical ectopia we showed co-existence of *C. trachomatis* infection. It is in concordance with the data of other authors: 46.9% reported by Giedrys-Kalemba *et al.* (1994) and 39.7% by Barnes *et al.* (1990). However, when analyzing cervical ectopia rate in women with and without *C. trachomatis* infection, we obtained similar results (24.3% and 28.7%, respectively).

Urogenital mycoplasmas are frequently isolated from clinical samples. We did not notice differences in frequency of colonization by urogenital mycoplasmas among women with chlamydial and non-chlamydial cervicitis. The ratio was 27.2% and 24.7%, respectively (Table I). Maeda *et al.* (2004) did not observe statistically significant differences in the frequency of isolation of mycoplasmas among NGU patients with- and without chlamydial infection. *U. urealyticum* was isolated much more often than *M. hominis,* which agrees well with the results of other authors (Keane *et al.*, 2000, Schlicht *et al.*, 2004). Schlicht *et al.* (2004) showed high prevalence of genital mycoplasmas among sexually active young women with cervicitis (54% for ureaplasmas and 26% for *M. hominis*). They also observed a high level (16%) colonization of healthy female volunteers by mycoplasmas. High level of mycoplasmal colonization in asymptomatic women was also reported by Keane *et al.* (2000): appropriatly 29% for *U. urealyticum* and 12% for *M. hominis*.

In our study we demonstrated 14% of GBS-positive swabs obtained from vaginal fornix. We did not observe any significant correlation between occurrence of *C. trachomatis* and GBS or urogenital mycoplas-

mas and GBS. Honig *et al.* (2002) did not demonstrate any correlation of vaginal colonization with GBS and chlamydial infection or other STIs. Frequency of isolation for these streptococci from the urogenital tract of healthy women was estimated to be 7% to 34% (Bayo *et al.*, 2002, Manning *et al.*, 2001).

In spite of long-time using of penicillins in the treatment of streptococcal infections GBS are still sensitive to this group of antibiotics. The sensitivity of GBS to penicillins and percentage of resistance to erythromycin (9.6%) and clindamycin (7.7%) in our study was similar to that reported by others (Stiller *et al.*, 2003, Weisner *et al.*, 2004).

Our study confirms high frequency of *C. trachomatis* infection among women with cervicitis in the region of Upper Silesia. However we demonstrated that *C. trachomatis* infection does not influence urogenital colonization by mycoplasmas and GBS.

Literature

Bakken I.J., F.E. Skjeldestad, T. Ovreness, S.A. Nordbo and G. Storvold. 2004. Prevalence of Chlamydia trachomatis by sexual behaviour among young women. *Tidsskr. Nor. Laegeforen.* **124**: 1633–1635.

Barnes R.C., B.P. Katz, R.T. Rolfs, B. Batteiger, V. Caine and R.B. Jones. 1990. Quantitative culture of endocervical Chlamydia trachomatis. *J. Clin. Microbiol.* **28**: 774–780.

Bayo M., M. Berlanga and M. Agut. 2002. Vaginal microbiota in healthy pregnant women and prenatal screening of group B streptococci (GBS). *It. Microbiol*. **5**: 87–90.

Choroszy-Król I. and J. Ruczkowska. 1994. The prevalence of *C. trachomatis* genitourinary tract infection 1986–1993. *Przegl. Epidemiol.* **48**:261–264.

Critchlow C., P. Wolner-Hanssen, D. Eschenbach, N. Kiviat, L. Koutsky, C. Stevens and K. Holmes. 1995. Gynecology. Determinants of cervical ectopia and cervicitis: Age, oral contraception, specific cervical infection, smoking and douching. *Am. J. Obstet. Gynecol.* **173**: 534–543.

Egger M., N. Low, G.D. Smith, B. Lindblom and B. Herrmann. 1998. Screening for chlamydial infections and the risk of ectopic pregnancy in a county in Sweden: ecological analysis. *BMJ*. **316**: 1776–1780.

Friedek D., A. Ekiel, Z. Chelmicki and M. Romanik. 2004. HPV, *Chlamydia trachomatis* and genital mycoplasmas infections in women with Low Grade Squamous Intraepithelial Lesions (LSIL). *Ginekol. Pol.* **75**: 457–463.

Giedrys-Kalemba S., E. Wydra, J. Hałasa and I. Bilska. 1994. The biocenosis of the vagina and infection by Chlamydia trachomatis. *Med. Dośw. Mikrobiol.* **46**: 73–77.

Honig E., J.W. Mouton and van der W.I. Meidem. 2002. The epidemiology of vaginal colonisation with group B streptococci in a sexually transmitted disease clinic. *Eur. J. Obstet. Gynecol. Reprod. Biol.* **105**: 177–180.

Keane F.E., B.J. Thomas, C.B. Gilroy, A. Renton and D. Taylor-Robinson. 2000. The association of Mycoplasma hominis, Ureaplasma urealyticum and Mycoplasma genitalium with bacterial vaginosis: observations on heterosexual women and their male partners. *Int. J. STD AIDS.* **11**: 356–360.

Maeda S.I., T. Deguchi, H. Ishiko, T. Matsumoto, S. Naito, H. Kumon, T. Tsukamoto, O. Shouichi and S. Kamidono. 2004. Detection of Mycoplasma genitalium, Mycoplasma hominis, Ureaplasma parvum (biovar 1) and Ureaplasma urealyticum (biovar 2) in patients with non-gonococcal urethritis using polymerase chain reaction-microtiter plate hybridization. *Int. J. Urol*. **11**: 750–775.

Manning S.D., M.D. Pearlman, P. Tallman, C.L. Pierson and B. Foxman. 2001. Frequency of antibiotic resistance among group G Streptococcus isolated from healthy college students. *Clin. Infect. Dis.* **33**: 137–139.

Miller W.C., C.A. Ford and M. Morris. 2004. Prevalence of chlamydial and gonococcal infections among young adults in the United States. *JAMA*. **291**: 2229–2236.

Millman K., C.M. Black, R.E. Johnson, W.E. Stamm, R.B. Jones, E. Hook, D.H. Martirn, G. Bolan, S. Tavare and D. Dean. 2004. Population-based genetic and evolutionary analysis of *Chlamydia trachomatis* urogenital strain variation in the United States. *J. Bacteriol.* **186**: 2457–2465.

Nunez-Troconis J.T. 1999. Mycoplasma hominis and Ureaplasma urealyticum in different gynecologic diseases. *Invest. Clin.* **40**: 9–24.

Rastawicki W., H. Kalota, M. Jagielski and R. Gierczyński. 2004. Comparison of Polymerase Chain Reaction Assay and Mycoplasma IST 2Test with culture for detection of Infections caused by Ureaplasma urealyticum and Mycoplasma hominis. *Med. Dosw. Mikrobiol.* **56**: 99–108.

Schlicht M.J., S.D. Lovrich, S. Sartin, P. Karpinsky, S.M. Callister and W.A. Agger. 2004. High prevalence of genital mycoplasmas among sexually active young adults with urethritis or cervicitis. Symptoms in La Crosse, Wisconsin. *J. Clin. Microbiol.* **42**: 4636–4640.

Stiller R.J., L. Padilla, R. Choudhary, T. Tinghitella and S. Laifer. 2003. Group B streptococcal antibiotic resistance patterns in pregnant women. *Conn. Med*. **67**: 323–326.

Weisner A.M., A.P. Johnson, T.L. Lamagni, E. Arnold, M. Warner, P.T. Heath and A. Efstratiou. 2004. Characterization of group B streptococci recovered from infants with invasive disease in England and Wales. *Clin. Infect. Dis.* **38**: 1203–1208.

Zbroch T., P. Knapp, E. Błonska, M. Kobylec and P. Knapp. 2004. Chlamydia trachomatis infection and Bacterial vaginosis in the aspect a life style and their impact on cervical lesions occurrence. *Ginek. Pol.* **75**: 538–544.