

The frequency of gonorrheal and chlamydial infections in Zanjanian women in 2013-2014

Behnaz Molaei¹ M.D., Farnaz Mohammadian¹ M.D., Maryam Eftekhari² M.D., Robabeh Hatami¹ M.D., Atefe Tirkan³ M.D. student, Mahsa Kiani³ M.D. Student.

1. Department of Obstetrics and Gynecology, Zanjan University of Medical Sciences, Zanjan, Iran.
2. Department of Obstetrics and Gynecology, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.
3. Zanjan University of Medical Sciences, Zanjan, Iran.

Corresponding Author:

Farnaz Mohammadian, Zanjan University of Medical Sciences, Azadi Ave., Zanjan, Iran.
Email: mohamadian@zums.ac.ir; Mohammadian_farnaz@yahoo.com
Tel: (+98) 9122413354

Received: 10 October 2015
 Revised: 21 July 2016
 Accepted: 4 December 2016

Abstract

Background: *Chlamydia trachomatis* and *Neisseria gonorrhoeae* are the most prevalent bacterial sexually transmitted diseases in women.

Objective: The purpose of this study was to investigate the prevalence of gonorrheal and chlamydial infections and determination of related risk factors in married women with vaginal discharge attending gynecological outpatient department (OPD) in Zanjan in 2013-2014.

Materials and Methods: In this cross sectional study, 100 married women aged 18-49 years with vaginal discharge were evaluated for signs and symptoms of gonococcal and chlamydial infections. Then cervical discharge samples and blood samples were collected from each subject for the detection of *Nisseria gonorrhea* and *Chlamydia trachomatis* by bacterial culture and serological tests, respectively.

Results: The overall prevalence of *Chlamydia trachomatis* and *Nisseria gonorrhoeae* were 16% and 4%, respectively. There was no significant relationship between the contraception methods, previous history of vaginal infections, previous history of urinary tract infections, number of coitus per week and self-reported symptoms (itching, burning, abdominal pain) with prevalence of *Nisseria gonorrhoeae* and *Chlamydia trachomatis*.

Conclusion: According to our results, the prevalence of gonococci infection in Zanjan was remarkable and relatively was higher than other parts of Iran, therefore it is necessary to put emphasis on education and further preventive and therapeutic programs.

Key words: Vaginal discharge, *Nisseria gonorrhea*, *Chlamydia trachomatis*, Prevalence.

This article extracted from M.D. thesis. (Atefe Tirkan, Mahsa Kiani)

Introduction

Sexually transmitted infections (STIs) are major public health concern worldwide, especially in developing countries due to limited resources and facilities to diagnose and treat them (1). And each year globally occurred around 500 million new curable STIs (2). According report of WHO in 2012, the estimated global prevalence of chlamydia and gonorrhea was 4.2% and 0.8% respectively (3).

Epithelial cells of the genital tract can be infected through *chlamydia trachomatis* as an obligate intracellular pathogen in infected women. Most of infected women have no symptoms and clear from infection spontaneously, but persistent infection can spread to their upper genital tract (4). *Chlamydia trachomatis* causes long term

complications such as pelvic inflammatory disease, chronic pelvic pain, ectopic pregnancy, and infertility (4). Gonorrhea is an infection due to *Neisseria gonorrhoeae* and usually involves mucosa of the cervix, urethra, endocervix, fallopian tubes, rectum, and throat (5, 6). Generally clinical symptoms for *gonorrhoeae* are not different from genital chlamydial infection and clinically are indistinguishable (4). *Chlamydia trachomatis* and *Neisseria gonorrhoeae* are asymptomatic in the most of infected women (7).

Prevalence of chlamydia and gonorrhea in the country is considerable, so that results of the national meta-analysis study showed that *C. trachomatis* has a high prevalence for women in Iran and ranged from 0 to 32.7% in different studies (8). Afrakhteh et al. showed that *Neisseria gonorrhoeae* was the fourth prevalent STI pathogens in females in Tehran

with the prevalence rate equal 4.91% (9). Literatures shows that Being young, female gender, number of sexual partners considered as risk factors of these two infections (10-12). But there is not enough evidence in regard of association between type of partners, use of contraceptive methods, and age of partners at first intercourse and increase risk of chlamydial (13).

Increasing the risk of persistent undiagnosed chlamydial and gonococcal infections may lead to complications such as pelvic inflammatory diseases, infertility, ectopic pregnancy, tubal factor infertility, chronic pelvic pain, cervical infection, premature delivery, and low birth weight (14-18). The prevention of the occurrence complications and curb the spread of infection in symptomatic and asymptomatic patients usually is depends on early and accurate diagnosis and also appropriate treatment of infection (19). Also early detection of these STIs because of their synergistic effect with HIV infection is extremely important (20).

For proposing the strategies to prevent and control these infections we needed to precise data collection focused on distribution of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* and demographic characteristics (21). "While high STI prevalence indicates frequent risky sexual practice and a poor provision or uptake of services, low STI prevalence reflects the improvement in provision of care services or change in risky behaviors"(22). Based on this evidence, it is essential to carry out such studies.

Therefore, the purpose of this study was to investigate the prevalence of *Chlamydia trachomatis* and *Neisseria gonorrhoeae*, and determination of related risk factors in married women with or without symptoms in Gynecological OPD in Zanjan in 2013-2014.

Materials and methods

In this Cross sectional (descriptive/analytic study), 100 married women, 18-49 year old who were suffering from vaginal discharge with or without symptoms (itching, irritation and abdominal pain) referred to the

Gynecology Clinic at Ayatollah Mousavi Hospital in Zanjan, west of Iran, during 2013-2014, were included in the study.

Virgin and pregnant women, women at the time of menopause, history of chronic diseases such as diabetes, kidney transplantation, immune system disorders, corticosteroid use, females with history of using vaginal cream, vaginal douching and sexual intercourse in the last week, as well as using oral antibiotic during the past 48 hr were considered as exclusion criteria's.

Data were gathered by a checklist. The checklist used in this study was including: data on demographic characteristics, body mass index (BMI), educational level, occupation, pregnancy status, vaginal and genital tract infections, and contraceptive method. Also, their symptoms such as local itching, burning, dysuria, abdominal pain, abnormal vaginal discharge, the amount of discharge (low, high, normal), discharge color (white, green, and yellow), and the smell of the discharge were listed.

Then cervical discharge and blood samples were collected from each participant for the detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* by bacterial culture and serological tests, respectively. The endocervix was first cleaned with a sterile cotton swab to remove mucous and exudates then samples were obtained from endocervix by a sterile applicator. Two cervical swabs for detecting of *Neisseria gonorrhoeae* were collected from participants by a gynecologist. One cervical swab was taken to prepare the smear on a sterile glass slide, and the remaining swab sample was placed into a test tube. Once the samples were obtained, they were transported to Laboratory at Ayatollah Mousavi Hospital in Zanjan, Iran.

For isolation and identification of *Neisseria gonorrhoeae*, cervical swab samples were directly inoculated in modified Thayer Martin Media (MTM) (MERCK Co., Germany) and they were maintained in 5% CO₂ at 37°C. Isolates were identified as *Neisseria gonorrhoeae* on the basis of colony morphology, Gram staining, the oxidase, catalase and carbohydrate utilization tests.

Gram staining was performed using Gram stain kit (Bahar Afshan Co., Iran). In this study the Enzyme Immune Assay (ELISA) method was used to assess chlamydial infection. 2 mL blood sample was taken from each subject and was transferred to the laboratory for separation of serum. Serum IgM and IgG antibodies against *Chlamydia* were measured by enzyme-linked immunosorbent assay (ELISA) method (NOVA TEC kit, Germany).

Ethical consideration

The study protocol was approved by the Ethics Committee of Zanjan University of Medical Sciences, Zanjan, Iran. A written informed consent was taken from all participants.

Statistical analysis

The collected data were analyzed using the Statistical Package for Social Science (SPSS Inc., Chicago, version 11.5). Descriptive results were expressed as mean and standard deviation. Chi-square tests (χ^2), and Fisher exact test were used for analysis of categorical data. $P < 0.05$ was considered statistically significant.

Results

There were 100 women with vaginal discharge, with a mean \pm SD age of 33.05 \pm 7.97 years. BMI ranged from 18.37-37.17 kg/m² (mean \pm SD=26.78 \pm 18.37). Most women had been referred to the clinic in the second half of the menstrual cycle. As shown in table 1, only 10% of patients had higher education than diploma and 23% were illiterate. 17% of participants in the study did not have a history of pregnancy or childbirth. 61% of participants used withdrawal method of contraception. Abdominal pain was the common observed clinical symptom among them (62%).

The overall prevalence rate of Chlamydia trachomatis infection was 16% (16 women) in our participants. ELISA tests indicated that 16 women had positive antibodies' level in their

blood: 1 woman had anti-Chlamydia trachomatis IgM antibody, 12 had positive IgG antibody, and 3 women had both of them. *Neisseria gonorrhoeae* were detected in 4 (4%) participants. According to table II, there was no significant relationship between the contraception methods, history of vaginal infections, history of urinary tract infections, number of sexual intercourse weekly, and self-reported symptoms with the prevalence of *Neisseria gonorrhoeae* and *Chlamydia trachomatis* ($p > 0.05$).

Table I. Frequency of studied variables in 100 women with vaginal discharge attending gynecological OPD in Zanjan

Variables	n	%
Education of women		
Illiterate	23	23
Under diploma	40	40
Diploma and associate degree	27	27
BA and higher	10	10
Age group		
<30 year	58	58
≥ 30 year	42	42
Number of pregnancy		
0	17	17
1	25	25
2	30	30
$3 \leq$	28	28
Contraceptive method		
Pill	15	15
DMPA	4	4
IUD	4	4
Tubectomy or vasectomy	9	9
Condoms	7	7
Withdrawal method	61	61
Number of coitus weekly		
1-2	64	64
3-5	29	29
≥ 5	7	7
The clinical symptoms observed		
Itching	59	59
Burning	47	47
Abdominal pain	62	62
The amount of discharge		
Low	40	40
High	39	39
Normal	21	21
Discharge color		
White	49	49
Yellow	47	47
Green	4	4
Type of discharge		
Foamy	4	4
Clear	38	38
Thick	38	38
Cottage cheese	20	20
Unpleasant smell of the discharge	61	61

BA: Bachelors academic degree,
DMPA: Depomedroxyprogesterone acetate
IUD: Intra uterine device for contraception

Table II. Relation between studied variables and prevalence of *Neisseria gonorrhoeae* and *Chlamydia trachomatis*

Variables	Chlamydia negative	Chlamydia positive	p-value	Gonorrhoeae negative	Gonorrhoeae positive	p-value
History of UTI						
Yes	14 (16.7)	4 (25)	0.48	16 (16.7)	2 (50)	0.14
No	70 (83.3)	12 (75)		80 (83.3)	2 (50)	
history of vaginal infections						
Yes	58 (62.5)	10 (62.5)	0.77	65 (67.7)	3 (75)	1
No	26 (31)	6 (37.5)		31 (32.3)	1 (25)	
Contraceptive method						
Pill	13 (15.5)	2 (12.5)	0.98	14 (14.7)	1 (25)	0.31
DMPA	3 (3.6)	1 (6.3)		4 (4.2)	0	
IUD	4 (4.8)	0		3 (3.1)	1 (25)	
Tubectomy or vasectomy	8 (9.5)	1 (6.3)		9 (9.4)	0	
Condoms	6 (7.1)	1 (6.3)		7 (7.3)	0	
Withdrawal method	50 (50.5)	11 (68.8)		59 (61.5)	2 (50)	
Number of coitus weekly						
1-2	50 (59.5)	14 (87.5)	0.14	62 (64.5)	2 (50)	0.69
3-5	27 (32.1)	2 (12.5)		27 (28.1)	2 (50)	
≥ 5	7 (8.3)	0		7 (7.3)	0	
The clinical symptoms observed						
Itching	46 (54.8)	13 (81.3)	0.56	58 (60.4)	1 (25)	0.3
Burning	39 (46.4)	8 (50)	1	46 (47.9)	1 (25)	0.6
Abdominal pain	54 (64.3)	8 (50)	0.4	59 (61.5)	3 (75)	0.9

Discussion

This is a cross sectional study regarding investigate the prevalence of *Chlamydia trachomatis* and *Neisseria gonorrhoeae*, and determination of related risk factors in married women with vaginal discharge in Gynecological OPD in Zanjan. The overall prevalence of *gonorrhoea* infection in this study was 4%. Our results were consistent with the incidence reported by Afrasiabi *et al* using culture techniques in Kashan, Iran and Akya *et al* using PCR techniques in Kermanshah, Iran for *Neisseria gonorrhoeae* infection (21, 23).

In the present study, the prevalence of gonococci infection was relatively higher than some of national studies. For example in a study conducted by Haghghi *et al* in Sabzevar, Iran, the reported incidence of *Neisseria gonorrhoeae* was 1.25% (24). In another study were done by Bakhtiari *et al* in the north of Iran, the incidence of *Neisseria gonorrhoeae* was 0.2% (25). In one study in Zanjan in 2009, 308 vaginal samples from women were evaluated. They reported that the prevalence of *Neisseria gonorrhoeae* was 0.9% (26). Similar studies have had different results at the international level, for example consistent with our results in a study in health clinic attendees complaining of vaginal discharge in Bangladesh, the prevalence of *Neisseria gonorrhoeae* has been reported 3.8%, a higher rate of gonococcal prevalence

(4.0%) was reported in women of Brazil (27, 28).

Also one study in Turkey that used PCR reported a *Neisseria gonorrhoeae* infection incidence of 3.4%, which was relatively similar to our findings (29). While some studies reported lower rate than our study, the prevalence of *Neisseria gonorrhoeae* was 1.9% in the symptomatic population in Italy (30). Furthermore Farraj and his colleagues in Palestine used PCR to examine 213 endocervical samples and reported a *Neisseria gonorrhoeae* infection incidence of 1.4% (31). Or the prevalence rates of *Neisseria gonorrhoeae* in urine samples study of Ramos de Lima *et al* were 0.7% by PCR. In this Brazilian study because the type of specimen used for diagnosis, prevalence rate was lower than our study. Although nucleic acid amplification tests are highly appropriate for the detection of genital gonococcal infection, In addition PCR test for demonstration of *Neisseria gonorrhoeae* detection in urine samples has lower sensitivity (5). By contrast in Australia, baseline prevalence was 9.5% for *Neisseria gonorrhoeae*, which was higher than our result (32). "The reasons for discrepancy in gonorrhoea rates are not well understand, while probably include differences in access to health services and utilization, sample size, geographic clustering of populations, other interrelated social and economic factors, and sexual partner choices along both

socioeconomic and racial lines "(33). *Gonorrhoea* is a major public health concern globally that requires immediate international public health resources and attention (34).

while the most of symptomatic infected women were remained untreated because they did not seek care, and 50% of asymptomatic infected women were undetected and untreated (35). Thus, the control of this disease will require strategies such as screening and mass treatment, in addition to improved clinical services for patients with symptoms (36). The isolation of *Neisseria gonorrhoeae* from endocervical specimens by culture is the suggested method for isolation of gonorrhoea in women (21).

The overall prevalence of chlamydia infection in this study was 16%. This result suggests a high frequency rate of *Chlamydia trachomatis* infection in Zanjan, Iran. In our study the detection rate of *Chlamydia trachomatis* was relatively higher than the results of the other studies in Iran. Akya *et al* reported a 3.1% incidence of *Chlamydia trachomatis* by PCR in Kermanshah, Iran (23). In the United States the incidence of *Chlamydia trachomatis* in young adults has been reported around 10%, which was lower than our findings (37).

The prevalence of *Chlamydia trachomatis* in our study (16%) is higher than sexually active young Brazilian women (7.4% and 13.5%) that were examined in two separate study (38, 39), and also in similar studies in Greenland, USA, Norway, Canada and Uganda with the prevalence of chlamydia infection 7%, 8%, 7.2%, 9.3% and 6% respectively (40-44). However differences in the prevalence of *chlamydia* and *gonorrhoea* in different studies may be due to the use of different diagnostic methods, differences in sample size and different socio-cultural conditions.

In our study the prevalence of infection for both organisms was higher in women aged younger than 30 year (58% vs. 42%). Similarly, some of studies showed that the younger aged women are at higher risk for *Chlamydia trachomatis* and *Neisseria gonorrhoeae* (5, 21, 23). It is obvious that the younger aged women have higher sexual promiscuity therefore they are at higher risk for STD (45). According to findings of this

study bacterial infection has higher rate among women with use of withdrawal method. These results are similar to the results reported by Yirenya-Tawiah *et al* and Akya *et al* (23, 46). As shown in the results of this study, only 7% of women have used barrier methods, it seems that the high prevalence of chlamydia and gonorrhoea among women in this study partly can be explained through it.

Our findings did not show any relation between self-reported symptoms and positive laboratory tests for chlamydial and *gonorrhoeae*. These results was concordance with the study performed by Yirenya-Tawiah *et al* in Ghana, Nevertheless, they observed only a small percentage of the STI positive cases to report the presence of symptoms related to STIs (46). In a study was done in Batswana, many *Chlamydia trachomatis* and *Neisseria gonorrhoeae* infected individuals were asymptomatic (47). In another study by Akya *et al*, the difference in the rate of infection for both *Chlamydia trachomatis* and *Neisseria gonorrhoeae* among symptomatic and asymptomatic women was not statistically significant, indicating the silent infections of these bacteria among women (23). Consistent with our finding, Wasserheit *et al* in a meta-analysis study demonstrated that only 28% of women with *Chlamydia trachomatis* and *Neisseria gonorrhoeae* infection were reported to show symptom of vaginal discharge (48). Wasserheit *et al* also found that in 92% of women with vaginal discharge were not infected to gonococcal and chlamydial infection. She concludes that syndromic approach should not be used as a screening procedure for case finding of gonococcal and chlamydial (48).

Limitations

The study presents some limitations; clinical data such as sexual behavior and number of sexual partners was not consistently recorded and were thus not included into the study results. A number of patients were not working to obtain blood samples and leading to longer process of collecting samples. To identify high risk patients of transmission STD, we required a detailed biography about sexual relations, multiple sexual partners, type and history of marriage, that none of the patients cooperated

to complete information. Also patients did not give us properly information about clinical signs of their intimate partner, these cases may be due to constraint issues related to face-to-face interviews. Future studies are needed to carefully record sexual partner and provide additional epidemiologic information.

Conclusion

The results of this study revealed the prevalence of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in women suffering from vaginal discharge is relatively high. It may be because there is no clinic for sexually transmitted diseases in Zanjan, Iran and patients with urogenital infections referred to gynecology which can increase the positive results. According to results obtained through laboratory tests, the prevalence of gonococcal and chlamydial infection in Zanjan is relatively high, which makes it necessary to put emphasis on education and further preventive and therapeutic programs. It seems that the prevalence of asymptomatic chlamydial and *gonorrhoeae* infection in women to be high, therefore focus on clinic-based testing is inadequate.

Acknowledgments

The data in this paper is the result of a medical student doctoral degree thesis supported and funded by Zanjan University of Medical Sciences, Zanjan, Iran.

Conflict of interest

The authors have no conflict of interest to report.

References

- Lamb ML, Douglas JM Jr. Sexually transmitted infections. In: Detels R, Beaglehole R, Lansang MA, et al. Oxford Textbook of Public Health. 5th Ed. Oxford, England: Oxford University Press; 2009.
- WHO. Sexually transmitted infections (STIs): updated fact sheet. 2013. <http://www.who.int/mediacentre/factsheets/fs110/en/index.html>.
- Newman L, Rowley J, Vander Hoorn S, Wijesooriya NS, Unemo M, Low N, et al. Global estimates of the prevalence and incidence of four curable sexually transmitted infections in 2012 based on systematic review and global reporting. *PLoS one* 2015; 10: e0143304.
- Dean D. Chlamydia trachomatis today: treatment, detection, immunogenetics and the need for a greater global understanding of chlamydial disease pathogenesis. *Drugs Today* 2009; 45(Suppl.): 25.
- de Lima YAR, Turchi MD, Fonseca ZC, Garcia FLB, e Cardoso FAdB, da Guarda Reis MN, et al. Sexually transmitted bacterial infections among young women in Central Western Brazil. *Int J Infect Dis* 2014; 25: 16-21.
- Johnson RE, Newhall WJ, Papp JR, Knapp JS, Black CM, Gift TL, et al. Screening tests to detect Chlamydia trachomatis and Neisseria gonorrhoeae infections-2002. *MMWR Recomm Rep* 2002; 51: 1-22.
- Cecil JA, Howell MR, Tawes JJ, Gaydos JC, McKee KT, Quinn TC, et al. Features of Chlamydia trachomatis and Neisseria gonorrhoeae infection in male Army recruits. *J Infect Dis* 2001; 184: 1216-1219.
- Ahmadi MH, Mirsalehian A, Bahador A. Prevalence of genital Chlamydia trachomatis in Iran: a systematic review and meta-analysis. *Pathogens Global Health* 2015; 109: 290-299.
- Afrakhteh M, Beyhaghi H, Moradi A, Hosseini SJ, Mahdavi A, Giti S, et al. Sexually transmitted infections in Tehran. *J Family Reprod Health* 2008; 2: 123-128.
- Einwalter LA, Ritchie JM, Ault KA, Smith EM. Gonorrhea and chlamydia infection among women visiting family planning clinics: Racial variation in prevalence and predictors. *Perspect Sex Reprod Health* 2005; 37: 135-140.
- CDC. Sexually Transmitted Disease Surveillance: Centers for Disease Control and Prevention; 2001.
- Opoku BK, Sarkodie Y. Prevalence of genital Chlamydia and Gonococcal infections in at risk women in the Kumasi metropolis, Ghana. *Ghana Med J* 2010; 44: 21-24.
- Navarro C, Jolly A, Nair R, Chen Y. Risk Factors for Genital Chlamydial Infection: A Review. *Canadian J Infect Dis Med Microbiol* 2002; 13: 195-207.
- Banikarim C, Chacko MR, editors. Pelvic inflammatory disease in adolescents. Seminars in pediatric infectious diseases; 2005: Elsevier.
- Chacko MR, Wiemann CM, Smith PB. Chlamydia and gonorrhea screening in asymptomatic young women. *J Pediatr Adolesc Gynecol* 2004; 17: 169-178.
- Pellati D, Mylonakis I, Bertoloni G, Fiore C, Andrisani A, Ambrosini G, et al. Genital tract infections and infertility. *Eur J Obstet Gynecol Reprod Biol* 2008; 140: 3-11.
- Manavi K. A review on infection with Chlamydia trachomatis. *Best Pract Resh Clin Obstet Gynaecol* 2006; 20: 941-951.
- Mylonas I. Female genital Chlamydia trachomatis infection: where are we heading? *Arch Gynecol Obstet* 2012; 285: 1271-1285.
- Manhart LE, Broad JM, Golden MR. Mycoplasma genitalium: should we treat and how? *Clin Infect Dis* 2011; 53 (Suppl.): S129-S42.
- Wassrheit J. Interrelationships between human immunodeficiency virus infection and other

- sexually transmitted diseases. *Sex Transm Infect* 1992; 19: 61-76.
21. Afrasiabi S, Moniri R, Samimi M, Mousavi SG. The frequency of *Neisseria gonorrhoeae* endocervical infection among female carrier and changing trends of antimicrobial susceptibility patterns in Kashan, Iran. *Iran J Microbiol* 2014; 6: 194.
 22. Sopheab H, Morineau G, Neal JJ, Saphonn V, Fylkesnes K. Sustained high prevalence of sexually transmitted infections among female sex workers in Cambodia: high turnover seriously challenges the 100% Condom Use Programme. *BMC Infect Dis* 2008; 8: 167.
 23. Akya A. The frequency of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* infections among women in Kermanshah, Iran. *Asian Biomed* 2013; 7.
 24. Haghghi Hasanabad M, Bahador A, Mohammadzadeh M, Haghghi F. Prevalence of *Chlamydia Trachomatis*, *Neisseria Gonorrhoeae* and *Ureaplasma Urealyticum* in Pregnant Women of Sabzevar-Iran. *Sex Transm Infect* 2013; 89: 233.
 25. Bakhtiari A, Firoozjahi A. *Chlamydia trachomatis* infection in women attending health centres in Babol: prevalence and risk factors. *East Mediter Health J* 2007; 13: 1124-1131.
 26. Baghchesaraei H, Amini B, Hossaini M. Prevalence of infection with *Neisseria gonorrhoeae* and *Chlamydia trachomatis* in women visitors of gynecology and obstetrics clinics in Zanjan Province of Iran. *Afr J Microbiol* 2011; 5: 2447-2450.
 27. Rahman S, Garland S, Currie M, Tabrizi S, Rahman M, Nessa K, et al. Prevalence of *Mycoplasma genitalium* in health clinic attendees complaining of vaginal discharge in Bangladesh. *Int J STD AIDS* 2008; 19: 772-774.
 28. Rodrigues M, Fernandes P, Haddad J, Paiva M, Souza MDCM, Andrade T, et al. Frequency of *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Mycoplasma genitalium*, *Mycoplasma hominis* and *Ureaplasma* species in cervical samples. *J Obstet Gynaecol* 2011; 31: 237-241.
 29. Eksi F, Dikensoy E, Gayyurhan ED, Balci I, Balat O, Karsligil T, et al. The prevalence of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in the endocervical swab specimens of symptomatic, asymptomatic and infertile women in Turkey. *Arch Gynecol Obstet* 2011; 283: 1349-1355.
 30. Trevisan A, Mengoli C, Rossi L, Cattai M, Cavallaro A. Epidemiology of reproductive tract infections in a symptomatic population of North-East of Italy. *Minerva Ginecol* 2008; 60: 135-142.
 31. Farraj MA, Abusada GM, Saleem AAM, Joaidi AY, Radad RM, Atrash HN, et al. Detection of *Neisseria gonorrhoeae* in Palestinian women using polymerase chain reaction. *Asian Biomed* 2010; 4: 637-640.
 32. Silver BJ, Guy RJ, Wand H, Ward J, Rumbold AR, Fairley CK, et al. Incidence of curable sexually transmissible infections among adolescents and young adults in remote Australian Aboriginal communities: analysis of longitudinal clinical service data. *Sex Transm Infect* 2014; 0: 1-7.
 33. Gueye SB, Diop-Ndiaye H, Gningue A, Ndiaye O, Mbengue AS, Gaye-Diallo A, et al. Performance of the Abbott Real Time CT/NG assay in urines and cervico-vaginal samples from Senegal. *J Infect Develop Countries* 2014; 8: 898-903.
 34. Unemo M, Golparian D, Shafer WM. Challenges with gonorrhoea in the era of multi-drug and extensively drug resistance—are we on the right track? *Exp Rev Anti-Infect Ther* 2014; 12: 653-656.
 35. Wilkinson D, Karim SA, Harrison A, Lurie M, Colvin M, Connolly C, et al. Unrecognized sexually transmitted infections in rural South African women: a hidden epidemic. *Bullet World Health Organiz* 1999; 77: 22-28.
 36. Grosskurth H, Mayaud P, Mosha F, Todd J, Senkoro K, Newell J, et al. Asymptomatic gonorrhoea and chlamydial infection in rural Tanzanian men. *BMJ* 1996; 312: 277-280.
 37. Chernesky M, Jang D, Portillo E, Chong S, Smieja M, Luinstra K, et al. Abilities of APTIMA, AMPLICOR, and ProbeTec assays to detect *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in PreservCyt ThinPrep Liquid-based Pap samples. *J Clin Microbiol* 2007; 45: 2355-2358.
 38. Barcelos MRB, Vargas PRMd, Baroni C, Miranda AE. Genital infections in women attending a Primary Unit of Health: prevalence and risk behaviors. *Revista Brasileira de Ginecologia e Obstetrícia* 2008; 30: 349-354.
 39. Fernandes AMdS, Daher G, Nuzzi RXdP, Petta CA. *Chlamydia trachomatis* and *Neisseria gonorrhoeae* among women in a family planning clinic. *Revista Brasileira de Ginecologia e Obstetrícia* 2009; 31: 235-240.
 40. Datcu R, Gesink D, Mulvad G, Montgomery-Andersen R, Rink E, Koch A, et al. Vaginal microbiome in women from Greenland assessed by microscopy and quantitative PCR. *BMC Infect Dis* 2013; 13: 480.
 41. Gravningen K, Simonsen GS, Furberg A-S, Wilsgaard T. Factors associated with *Chlamydia trachomatis* testing in a high school based screening and previously in clinical practice: a cross-sectional study in Norway. *BMC Infect Dis* 2013; 13: 361.
 42. Lambert G, Haley N, Jean S, Tremblay C, Frappier J-Y, Otis J, et al. Sexual health of adolescents in Quebec residential youth protection centres. *Can J Public Health* 2013; 104: 216-221.
 43. Wiesenfeld HC, Lowry DL, Heine RP, Krohn MA, Bittner H, Kellinger K, et al. Self-collection of vaginal swabs for the detection of *Chlamydia*, gonorrhoea, and trichomoniasis: opportunity to encourage sexually transmitted disease testing among adolescents. *Sex Transm Dis* 2001; 28: 321-325.
 44. Rassajo E-B, Kambu F, Tumwesigye MN, Tenywa T, Darj E. Prevalence of sexually transmitted infections among adolescents in Kampala, Uganda, and theoretical models for

- improving syndromic management. *J Adolescent Health* 2006; 38: 213-221.
45. Norman J. Epidemiology of female genital Chlamydia trachomatis infections. *Best Pract Res Clin Obstet Gynaecol* 2002; 16: 775-787.
46. Yirenya-Tawiah D, Annang TN, Apea-Kubi KA, Lomo G, Mensah D, Akyeh L, et al. Chlamydia Trachomatis and Neisseria Gonorrhoeae prevalence among women of reproductive age living in urogenital schistosomiasis endemic area in Ghana. *BMC Res Notes* 2014; 7: 349.
47. Romoren M, Sundby J, Velauthapillai M, Rahman M, Klouman E, Hjortdahl P. Chlamydia and gonorrhoea in pregnant Batswana women: time to discard the syndromic approach? *BMC Infect Dis* 2007; 7: 27.
48. Wasserheit JN. The significance and scope of reproductive tract infections among Third World women. *Int J Gynecol Obstet* 1989; 30: 145-168.