Diploma Thesis

Epidemiological characteristics of the germ spectrum of urogenital infections in patients of the STD – outpatient clinic Graz May 2006 – May 2011

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Graz, 16.05.2014

Statutory Declaration

I declare that I have authored this thesis independently, that I have not used other than the declared sources and resources, and that I have explicitly marked all material which has been quoted either literally or by content from the used sources.

Graz, am 16.05.2014 *Georg Streitmayer*

Principle of Equality

In order not to disrupt the readability of this paper by a constant mentioning of both sexes, either a gender – neutral wording or only one of the two sexes is used. However, I want to emphasize explicitly that in either case, of course, both of the sexes are meant.

Note of thanks

At this point I would like to thank all those who have contributed through their professional and personal support to the success of this thesis.
I especially thank my supervisor Dr. Peter Komericki for providing the interesting topic of this thesis and the friendly helpfulness, he showed me.

I would like to use this opportunity to express my gratitude to my family, especially my parents, who not only allowed me my studies, but encouraged my ideas, gave me the opportunity to gather my own experiences and spend so much time abroad. Thank you so much for supporting me on what seemed to be an endless journey to finish this thesis.

Last, but not least I owe a big thank you to my friends for wonderful years in Graz and more to come.

Zusammenfassung

Hintergrund: Die Prävalenz von Chlamydia trachomatis, Neisseria gonorrhoeae, Ureaplasma urealyticum, Mycoplasma hominis und Trichomonas vaginalis in der Patientenpopulation der STD - Ambulanz der Universitätsklinik für Dermatologie und Venerologie Graz wurde statistisch bislang nicht erhoben und ausgewertet.

Ziel: Das Ziel dieser Arbeit ist es, die Prävalenz von sexuell übertragbaren Krankheitserregern und ihre Verteilung, bei den Patienten der STD - Ambulanz Graz zu untersuchen.

Methoden: Diese retrospektive Studie umfasste die Sammlung von Daten aller Patienten die auf Chlamydia trachomatis, Neisseria gonorrhoeae, Ureaplasma urealyticum, Mycoplasma hominis und Trichomonas vaginalis in der Zeit von 05/2006 bis 05/2011 in der STD -Ambulanz der Abteilung für Dermatologie und Venerologie Graz getestet wurden. Die folgenden Parameter wurden erhoben: Alter, Geschlecht und Infektionsstatus für die einzelnen Keime. Nach Abschluss der Sammlung aller Daten wurden diese anonymisiert und statistisch mit Microsoft Excel und SPSS ausgewertet.

Basierend auf den drei erwähnten Parametern (Alter, Geschlecht, Infektionsstatus) wurden zwei verschiedene Beobachtungzeiträume und daher zwei unterschiedliche Testpopulationen erstellt. Alle gesammelten Daten innerhalb des Zeitraums von 16.05.2006 bis 26.05.2011 wurden für die Auswertung der Parameter Alter und Geschlecht verwendet. Um die statistische Vergleichbarkeit der Infektionszahlen für die einzelnen Erreger im Jahresverlauf zu gewährleisten wurden für den Parameter Infektionsstatus nur die Jahre 2007 bis 2010 (jeweils volle 12 Monate) berücksichtigt.

Ergebnisse: Insgesamt wurden 1.669 Patienten, davon 1.159 (69%) Männer und 510 (31%) Frauen untersucht. Das Durchschnittsalter der Patienten lag bei 36.1 Jahren. Bei 39.4% (n = 657) der Patienten wurde mindestens eines der gesuchten Pathogene nachgewiesen. Von diesen 657 positiv getesteten Patienten waren 431 (n = 66%) männlichen und 226 (n = 34%) weiblichen Geschlechts.

Das Durchschnittsalter bei Patienten, die positiv auf mindestens einen der gesuchten Krankheitserreger getestet wurden, lag bei 31.8 Jahren. Der häufigste Erreger in der Studienpopulation zwischen 2007 - 2010 sowohl bei männlichen als auch bei weiblichen Patienten, war U. urealyticum (n = 319/61.8%), gefolgt von C. trachomatis (n = 108/20.9%), N. gonorrhoeae (n = 74/14.3%), M. hominis (n = 11/2.1%), und T. vaginalis (n = 4/0.8%). Die STD - Ambulanz Graz wurde am häufigsten von Patienten im Alter von 20 bis 29 Jahren aufgesucht. 392 Männer (33.8%) und 159 Frauen (31.1%) waren Teil

dieser Altersgruppe. Mit zunehmendem Alter (> 30) nahm die Häufigkeit der Konsultationen bei beiden Geschlechtern kontinuierlich ab.

Fazit: Aus den Ergebnissen der statistischen Auswertung der Daten der Grazer STD-Ambulanz läßt sich ableiten, dass sich die Prävalenz von C. trachomatis aber auch N. gonorrhoeae im untersuchten Zeitraum auf einem gleichbleibenden Niveau befindet, während die Infektionszahlen für U. urealyticum und M. hominis rückläufig sind. Aufgrund der geringen Prävalenz von T. vaginalis in der Patientenpopulation ist keine definitive Aussage möglich.

Abstract

Background: The prevalence of Chlamydia trachomatis, Neisseria gonorrhoeae, Ureaplasma urealyticum, Mycoplasma hominis and Trichomonas vaginalis in the patient population of the STD outpatient clinic of the Department of Dermatology and Venereology Graz has not yet been collected and statistically analyzed.

Objective: The goal of this thesis is to asses the prevalence of above mentioned pathogens among patients of the STD outpatient clinic Graz.

Methods: This retrospective study involved the collection of data of all patients tested for Chlamydia trachomatis, Neisseria gonorrhoeae, Ureaplasma urealyticum, Mycoplasma hominis and Trichomonas vaginalis in the period of time from 05/2006 to 05/2011 at the STD outpatient clinic of the Department of Dermatology and Venereology Graz. The following parameters were collected: age, sex and infection status. After completing the collection, all data were anonymized and analyzed statistically via Microsoft Excel and SPSS. Based on the three appointed parameters (age, sex, infection status) two different observation periods and consecutively two test populations were established. All data collected within the period of time from 05/16/2006 to 05/26/2011 were used for the evaluation of the parameters age and sex. To ensure statistical comparability of the collected data on the parameter infection status for each pathogen over the years only the years from 2007 to 2010 (full 12 months) were taken into account.

Results: In total 1669 patients, 1159 (69%) men and 510 (31%) women, were tested. The overall mean age of patients was at 36.1 years. Among all of the tested patients, 657 (39.4%) were tested positive for at least one of the pathogens of interest. Out of the 657 patients tested positive, 431 (n = 66%) were of male and 226 (n = 34%) of female gender.

The average age in patients tested positive for at least one pathogen was at 31.8 years. The most common pathogen to be found in the study population from 2007 to 2010 in both male and female patients was U. urealyticum (n = 319/61.8%), followed by C. trachomatis (n = 108/20.9%), N. gonorrhoeae (n = 74/14.3%), M. hominis (n = 11/2.1%), und T. vaginalis (n = 4/0.8%). The STD outpatient clinic Graz was most commonly frequented by patients between the age of 20 and 29 years. 392 men (33.8%) and 159 women (31.1%) were part of this age group. With increasing age (>30 years) the frequency of consultations at the Graz STD clinic in both sexes declined.

Conclusion: The results of the statistical evaluation of the data collected at the STD clinic Graz show that the prevalence of C. trachomatis and N. gonorrhoeae remains on a constant level, while U. urealyticum and M. hominis show declining numbers for the studied period

of time. A significant trend for T. vaginalis can not be determined, because of the small amount of infections detected.

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List of abbreviations

<	smaller/less than
>	bigger/more than
%	percentage
AIDS	Acquired Immune Deficiency Syndrome
ATP	Adenosine triphosphate
AOC	Adult chlamydial ophtalmia
C. trachomatis	Chlamydia trachomatis
DIF	Direct Immunofluorescence
DNA	Desoxyribonucleic acid
DGI	Disseminated gonococcal infection
ECDC	European Center for Disease Control
e.g.	example given
etc.	et cetera
ELISA	Enzyme linked immunosorbent assay
EU	European Union
EB	Elementary bodies
fig.	Figure
HBV	Hepatitis B virus
HIV	Human immunodeficiency virus
HPV	Human papillomavirus
IB	Inclusion bodies
ICN	Inclusion conjunctivitis of the newborn
IM	intramuscular
IV	intravenous
KAGES	Steiermärkische Krankenanstalten-Gesellschaft m.b.H.
LCR	Ligase chain reaction
LGTI	Lower genital tract infection
LGV	Lymphogranuloma venereum
MEDOCS	Eletronic communication- and information-network
MOMP	Major Outer Membrane Protein
MPC	Mucopurulent cervicitis
MSM	Men who have sex with men

M. hominis	Mycoplasma hominis
n.	Size of statistical sample
n.d.	not detected
N. gonorrhoeae	Neisseria gonorrhoeae
NAA	Nucleic acid amplification
NAH	Nucleic acid hybridization
NGNCU	Non-gonococcal non-chlamydial urethritis
NGU	Non - gonococcal urethritis
p.o.	per os
PCR	Polymerase chain reaction
PID	Pelvic inflammatory disease
PGU	Post - gonococcal urethritis
RB	Reticulate bodies
RNA	Ribonucleid acid
SARA	Sexually acquired reactive arthritis
SPSS	Statistical Product and Service Solutions
STD	Sexually transmitted diseases
STI	Sexually transmitted infections
tab.	Table
TMA	Transcription mediated amplification assay
T. vaginalis	Trichomonas vaginalis
UGTI	Upper genital tract infection
U.S. CDC	United States of America Center for Disease Control
U. urealyticum	Ureaplasma urealyticum
VD	Venereal diseases
WHO	World Health Organization

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1 Introduction

"One of the great misconceptions is that people who have STDs know they have them, ... That is absolutely incorrect" (1)

1.1 Sexually transmitted infections

1.1.1 Terminology or what are VDs, STDs, STIs?

On August 22nd 1945 the Austrian temporary government announced the first "Geschlechtskrankheitengesetz" (law of venereal diseases) after World War II. As defined by this law only Syphilis, Gonorrhea, Chancroid and Lymphogranuloma venereum (inguinale) were identified as venereal diseases (VD) and are still notifiable but only if the patient tries to elude therapy. (2)
Gradually over the years other diseases (e.g.: Chlamydia trachomatis, Herpes genitalis), which are primarily transmitted through sexual contact were included by linguistic usage and the term sexually transmitted diseases (STD) was installed. Nowadays using the term sexually transmitted infections (STI) became most usual, as this new definition emphasizes on the asymptomatic infection status of most of these diseases. In the following I will refer to the new definition STI as the most appropriate term. (3)
STIs are very heterogeneous. There are more than thirty different sexually transmissible pathogenic organisms, which vary from life - threatening to almost trivial, from acute to chronic and include bacterial, viral, fungal, protozoal infections as well as parasites. Some of them have been known for centuries others only recently discovered. What they all have in common, is their connection to the sexual sphere thus being not only a medical but also a social and political problem. (3,4)

1.1.2 Characteristics or what do STIs have in common?

According to the abbreviations meaning STIs are primarily transmitted during sexual contact, as humans are the only natural reservoir for STI pathogens. In addition to the sexual transmission, STIs can also be transmitted from mother to child during pregnancy or birth as well as through blood products and tissue transfer (e.g. intravenous drug use with shared needles). (3) Basically any sexually active person can be infected with an STI.

However, based on the number of entries and the epidemiological studies, some main affected groups can be defined by: (4)

- age: STI can occur in every age beyond puberty (under appropriate circumstances even before that), but the peak incidence is in early adulthood (25-30 years of age: the period of the highest sexual activity and most frequent changes of partners). (4)
- gender: Only under the age of 20 years female gender outweighs male, afterwards it is the other way around (average of 2.1 men : 1 woman). (4)
- demographics: The incidence of STIs is higher in cities than in rural areas and subsequently higher in metropolises than in small cities. (4)
- social standing: So called "Higher" social classes have fewer STIs than so called "lower" ones. As access to medical care is harder to get for the latter longer intervals between infection and treatment (duration of infectivity) occur. While the socially "higher" patients with venereal diseases are frequently treated by private physicians (where they do not supply to statistics). (4)
- profession and special circumstances: In addition to belonging to relevant segments of population (prostitutes, drug addicts, etc.), these are mainly groups of people forced to live under temporary sexual abstinence (e.g. military, naval crews, guest workers, long-distance truck driver, prison inmates). (4)
- sexual orientation: Male homosexuals (MSM), after all worldwide between 2 and 10% of male adults and thus a significant minority, pose a main affected group for genital contact infections, because of their sexual behavior and sexual practices. (4,5) Lesbian women are less concerned because their sexual practices are less invasive, and there is no deposition of potentially infectious semen. (4)

These main affected groups should not be equated with risk groups. By belonging to a group most affected, does not mean that a person behaves risky. Drawing such conclusions would be discriminatory and stigmatizing, because many members of the main stakeholder groups protect themselves and others. Strictly speaking there is no risk group, but only risky behaviour. (4)

Determining factors for spreading of STI in the population are: (4)

- de facto-infectivity of the pathogen
- average rate of partner change
- de facto-duration of infectivity

These factors are forming the following equation: (4)

$$R = ß \times c \times D$$

- R is the number of new infections during the observation period
- β stands for the transmission efficiency (transmission/ sexual intercourse)
- c is the average number of sexual partners
- D is the average duration of infectiousness (4)

Despite the diversity of pathogens and subsequent symptoms, their underlying pathological patterns are often similar. These patterns include genital ulcers and erosions with lymphatic transmission (e.g. syphilis, chancroid, lymphogranuloma venereum and granuloma inguinale). Other patterns are, not ascending (LGTI) and ascending (UGTI) infections of the cervix or urethra, caused by N. gonorrhoeae or C. trachomatis as well as vulvovaginitis (e.g. trichomonas vaginalis), virus localized lesions with recurrence probability (e.g. herpes genitalis), ectoparasitoses (e.g. scabies), systemic diseases (e.g. HBV, HIV), intestinal infections, and hepatitis A. (3,4)
Furthermore these agents share following characteristics: Their only host is man, they respond to physical and chemical stimuli and they are highly sensitive, especially to drought. They exclusively can be transferred by prolonged interaction in humid and hot environment or mechanical irritation. Many STI pathogens show tendencies to mixed or multiple infections, which in turn complicate treatment. Likewise an infection that has not been manifested clinically could be overlooked (e.g. lack of co-treatment of a not yet manifested syphilis by the use of gonorrhea therapy). Therefore screening for other STIs than the obvious one is crucial before starting treatment. (4)
Typical clinical signs that make you think of a STI in women are vaginal discharge, pelvic pain, irregular menstruation, and dyspareunia. In men, STIs cause urethral discharge, dysuria, balanitis and scrotal pain. Both men and women can suffer from genital ulcers, genital warts, rectal pain or discharge, mono or oligoathritis and conjunctivitis. These symptoms may be indicative for certain pathogens. (3)
Under certain circumstances sexually transmitted infections often remain silent and do not express themselves by obvious symptoms. If they stay asymptomatic or unnoticed it is especially problematic because then they remain untreated and therefore infectious. (6)

The consequences of untreated STIs can be severe. 40% of untreated bacterial STIs in females result in pelvic inflammatory disease, which in almost 30 - 40% leads to infertility due to post-infection tubal damage. (6) Moreover, sexually transmitted infections often cause complications during pregnancy, diseases of the newborn (e.g. infant blindness) and are important reasons for perinatal mortality, abortion and stillbirth. There is evidence that genital ulceration and genital warts as a result of STIs, increase both the transmission as well as acquisition of HIV infection. (6)

1.1.3 Epidemiology

1.1.3.1 Are STIs a global burden?

According to data of the WHO the world occurrence of STIs is only seconded by diarrheal diseases, lower respiratory diseases and malaria. Eight infections, syphilis, gonorrhea, chlamydia, trichomoniasis, genital herpes, HBV, and HPV, are accountable for the majority of STIs worldwide. (7-11)

The WHO estimates that 340 million new cases of STIs have occurred in 1999, with the largest growth rates in the region of South and Southeast Asia (151 million), followed by sub-Saharan Africa (69 million), Latin America and the Caribbean (38 million). (9)

By 2005 the WHO already assessed that syphilis, gonorrhea, chlamydia and trichomoniasis alone accounted for 448 million new cases of STIs in adults aged 15-49 years all over the world, which denotes a boost by 31.76% over the last 7 years. (7,8,12)

On that account the WHO launched a “Global Strategy for the prevention and control of sexually transmitted infections” to be implemented by 2006 to 2015 following the below mentioned objectives and components to reach four crucial points. (12)

(1) Reduction in STI-related morbidity and mortality;
(2) prevention of HIV through a cost-effective intervention;
(3) prevention of long-term sequelae of STIs, such as cancers, especially in women;
(4) reduction in adverse outcomes of pregnancy (in women infected with STIs) (12)

Objectives:

(a) Increase the commitment of national governments and national and international development partners for prevention and control;

(b) *Promote mobilization of funds and reallocation of resources, taking into account national prioritized results-oriented interventions that ensure aid effectiveness, ownership, harmonization, results and accountability;*

(c) *Ensure that policies, laws and initiatives related to provision of care are non-stigmatizing and gender-sensitive within the prevailing sociocultural context;*

(d) *Harness the strengths and capacities of all partners and institutions in order to scale up and sustain interventions for prevention and control.* (12)

Components:

- *availability or suitability of health-care services for priority target populations (e.g. adolescents and sex workers);*
- *diagnosis and treatment of asymptomatic infections;*
- *the syndromic approach for the management of abnormal vaginal discharge;*
- *management of sexually transmitted infections in sexual partners;*
- *attitudes of health-care providers;*
- *availability and reliability of data for planning purposes;*
- *global advocacy campaign to raise awareness and mobilize resources worldwide.* (12)

1.1.3.2 STIs in Europe or the recapturing of the old world

STIs are to be among the oldest and most common diseases of mankind. Changes in their numerical occurrence are mainly based on socio-economic conditions, the prevailing cultural and moral conception as well as on treatment options available. (3) STIs are playing an important role throughout history:

Naples, 1495. The young French king Charles VIII (1470-1498) tries to enforce his inheritance claims on Naples with a campaign. In spring of 1494 he moves with more than 30.000 men, most of them Spanish, Swiss, Dutch, French mercenaries, from Lyon via Italy to the south. King Ferdinand of Naples also recruites a mercenary army, also with many spaniards. Both kings not knowing that the outcome of the battle will be way more remarkable than the victory itself; because on both sides the Spanish mercenaries are the ones who carry the germ of the new disease that Columbus sailors brought back from their second expedition.

After the conquest of Naples by Charles VIII and weeklong celebrations, the army withdrew to the north. More and more mercenaries left the army on their way back, spreading the new disease all over Europe. (13,14)
On the topic of the ongoing debate about the history of syphilis, which usually is addressed with the simple question "America or Europe?" one can see that STIs have been, are and sadly will be a subject-matter to our society. "At the end only the devil will know", whether syphilis was really brought to Europe by Columbus, as Thomas Mann ironically expresses himself in Dr. Faustus about the "guests from the West Indies". (14,15)

During the post-war years after the first and second world war we noted a sharp rise of Gonorrhea and Syphilis among the German and Austrian population, which by the discovery of penicillin and the resulting new treatability of the classic bacterial venerea waned with a last peak of incidence during the "sexual revolution" in the 1970's. (3)
Since the late 1970's, the morbidity and mortality due to bacterial infection, such as syphilis and gonorrhea, declined significantly in Western Europe and other developed countries as a result of improved antibiotic therapy and increased use of condoms linked to the HIV epidemic. (16) This epidemic nevertheless started a wide spread debate about sexual habits and their changes to prophylaxis of infections. However, since the late 1990's, an increase in STIs in Western Europe can be observed. This is illustrated by the situation in England, Wales and Northern Ireland, where the number of reported new cases of gonorrhea and other STIs from 1995 to 2000 has more than tripled. (17,18) The situation is similar in Sweden and the Netherlands. The cases of Syphilis, Chlamydia infection, herpes genitalis and genital warts also increased in a number of cities and countries all over Europe mainly involving MSM, heterosexual prostitution contacts, young people and regular drug users. (17,19-21)
Reasons for the increasing number of infections are:

- growing population density
- increased immigration from areas of high infection rates
- enhanced mobility
- altered / decreasing sexual protective sexual behavior
- insufficient diagnosis and therapy services for vulnerable groups
- missing vaccines against STDs

The last two reasons are likely to be changed. There are already signs that some STDs in homosexual men are reclining due to changes in their sexual behavior. Vaccines against

certain infections (e.g. herpes simplex, gonorrhea, HIV) soon could be available for disposal. (6)

After the collapse of communism and the consequent crash of health system in Eastern European countries the VD's as well as HIV infections increased rapidly, partly 50 times more than before the breakdown. (4) As part of EU-enlargement to the East, border crossings and opportunities for residence and work choice were made much easier, consequently assisting the spread of infectious diseases. Therefore new challenges in the field of detection and prevention of STIs and HIV infection have been formed. (22)

In response the EU founded a project called BORDERNET between Germany, Poland, Austria, Slovakia, Italy and Slovenia. This project's task is to improve the existing health care structures in the various countries on the field of HIV/AIDS and STIs, and to network across borders. The aim of BORDERNET is to record the frequency and distribution of HIV and STDs in the participating EU border regions. Here, the main interest is less the collection of representative data but the early identification of epidemiological trends in certain population groups or regions as well as the collection of data on risk behaviors. First results of BORDERNET show a massive increase of Chlamydia infections. (22) According to data from BORDERNET prostitution represents the most significant risk for women to contract Chlamydia or Gonorrhea, for men it is homosexual behavior. (22)

In 2008, epidemiological data showed that with more than 335 000 cases reported, the Chlamydia infection is the most common bacterial STI in the EU. The reporting rates have more than doubled over the last 10 years. (23) This increase is largely due to measures such as screening and surveillance programs that have been taken in by the member states to improve the diagnosis and reporting of chlamydial infections. More than three quarters of all cases are diagnosed in young adults under 25. (23) Since the infection often is asymptomatic the actual number of cases might be even higher. Given the number of cases and the decline in fertility that occurs in some women, chlamydial infections constitute a relevant and growing burden of disease for individuals as well as for the entire healthcare systems. (23) Substantially lower rates are reported for gonorrhea and syphilis, which seem to be on a constant level. Between 2006 and 2009 there was a slight decrease (9%) in the total number of reported cases of both diseases. Higher rates were reported from the United Kingdom, the Netherlands and the Baltic states. (23)

In earlier years it was thought that the cause of the persistence of STIs despite all medical and legal efforts lay within the endemic occurrence (reservoirs) in latently infected individuals. Today there is a more dynamic picture: The persistence is explained by

constantly running small epidemics (infection chains) that wavelike pass through the risk groups. Only the peaks of these waves are discovered. Their broad base corresponds to asymptomatic infections or infections in the period of incubation. (4)

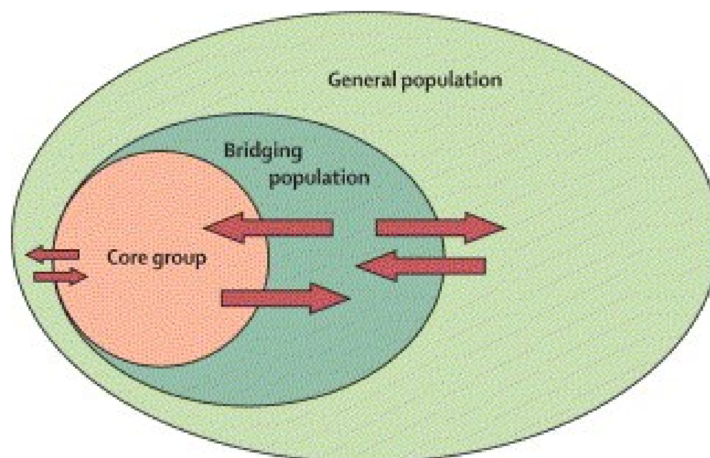


Figure 1 – Transmission dynamics of STI at the population level (24)

The goal of this thesis is to asses the prevalence of potential sexually transmitted pathogens, among patients of the STI – clinic Graz. Only pathogens detectable after urethral, cervical and vaginal smear got included in the study.

1.2 Chlamydia trachomatis

1.2.1 Etiopathogenesis

Chlamydiae, first discovered in 1907 by Halberstadt and Prowazek, were traditionally divided into three Chlamydia types: (25-28)

a. chlamydia trachomatis
b. chlamydia psittaci
c. chlamydia pneumoniae

Nowadays they are divided into the genera Chlamydia (a) and Chlamydophila (b, c) in the family of Chlamydiaceae. Chlamydia bacteria are very small, gram-negative and obligate intracellular living microorganisms with parasitic qualities. They enter the human body via tiny cracks in the mucous membrane. Since they are themselves incapable of synthesizing high-energy compounds (ATP) they relate on their host cells to provide for them. In order to reproduce themselves, Chlamydia bacteria rely on a life cycle way more complex than in free-living bacteria. The reproduction cycle is based on two distinct forms of bacterial development. (25-28)

- Extracellular, infectious elementary bodies (EB)
- Intracellular, replicative reticulate bodies (RB)
- Intracellular, inclusion bodies (IB)

EB attach themselves with specific molecules to receptors on susceptible target cells (epithelial cells) and are absorbed by these cells via endocytosis. Within eight hours EB convert into metabolically active RB, which subsequently reproduce themselves. This results in one or more of endosome membrane surrounded IB, which may contain thousands of Chlamydia particles (EB and RB) and occupy a large part of the infected host cell. After 24 to 36 hours RB are formed back into EB. Approximately 48 hours after the beginning of the reproduction cycle Chlamydia particles are eventually released by lysis (cytopathic effect) of the host cell or as an alternative parasitic way by exocytosis (the cell remains viable and able to divide and generates infected daughter cells). (25-28)

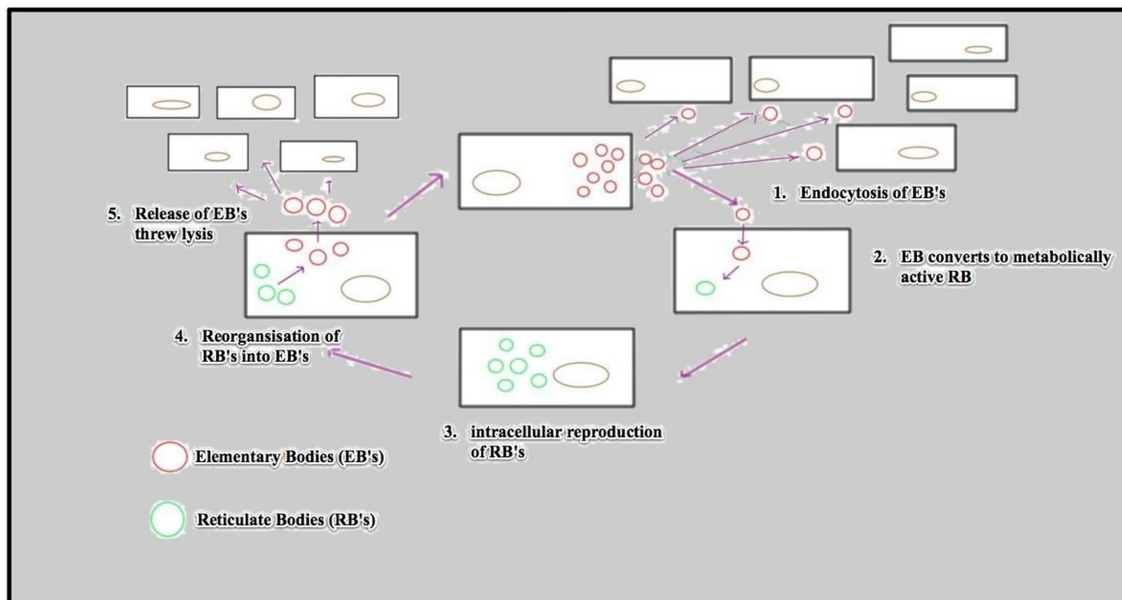


Figure 2 – Schematic representation of the replication cycle of Chlamydia

As an obligate intracellular bacterium Chlamydia trachomatis has a tropism for epithelia of the urogenital tract, the rectum and the conjunctiva. All in all there are 15 serotypes defined by different protein antigens, designated by the letters A to C, D to K and L1 to L3 in 3 groups. The different serotypes in a group share the same pathogenicity and are associated with certain medical conditions as shown in figure 3. (25-28)

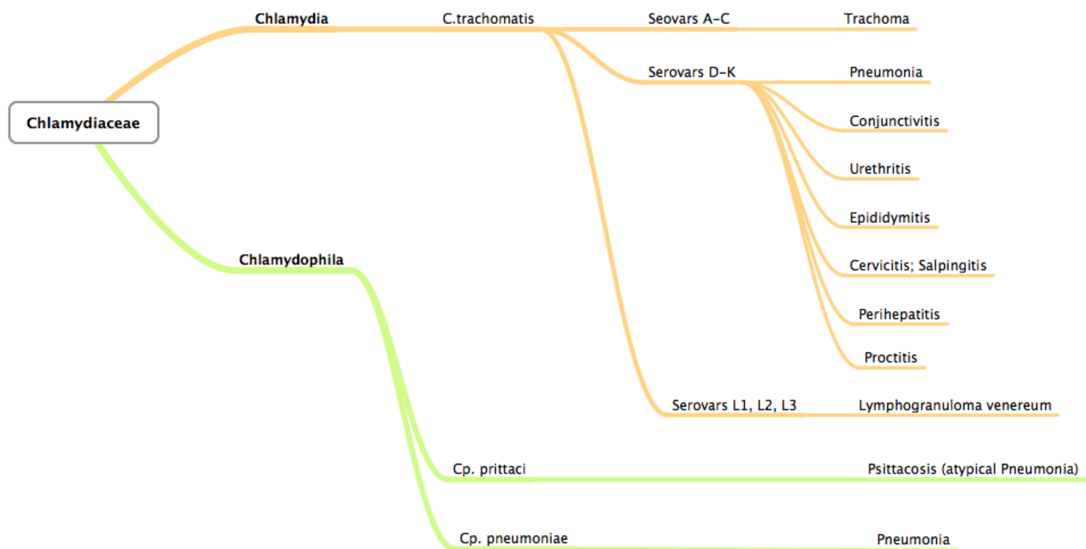


Figure 3 – Different characteristics of Chlamydiaceae

C. trachomatis is the most common STI in Europe. The infection is more common in women than in men and 75% of the cases is described in young adults (15-24 years). (29)

1.2.2 Path of infection

Serovars A - C are transmitted by smear infections of infectious eye secretions. Flies are important as vectors of trachoma. A transfer of serovars D-K and L1-L3 is possible only through sexual contact and perinatally, because humans are the only virus reservoir. After incubation the exact duration of infectiousness cannot be specified due to asymptomatic forms of the infection. In any case C. trachomatis looses its infectivity within 48 hours. (25-28)

1.2.3 Clinical Pathology

The clinical picture is a result of cell destruction and inflammatory host responses. The released EB can be transported by lymph or blood to close or distant situated cells from the site of infection and penetrate them. While the serotypes D - K of C. trachomatis are limited to the column and transitional epithelium and typically provoke "catarrhal" (serous - mucous) secretion with mild inflammation and mild general symptoms, the serotypes L1 - L3 can cause a systematic disease (LGV). Thus the clinical picture of an infection with C. trachomatis depends on the site of the infection and is a counterpart to gonorrhea. It differs from gonorrhea with its tendency to mild but - untreated - chronic - relapsing course. (25-28)

1.2.3.1 Genital infection in men with C. trachomatis serovars D-K

Primarily the epithelium of the distal urethra is infected. From there, the infection can spread over the entire urethra and sometimes over the vasa deferentia to the tubules of the epididymis. (25-28)

- Urethritis:

  C. trachomatis causes 30% of all cases of urethritis in men. Arising from times when only gonococci were assumed to be the cause of urethritis the following terms where used whenever no gonococci could be found: non-specific urethritis, non - gonococcal urethritis (NGU) or post – gonococcal urethritis (PGU). After an incubation period of one to three weeks patients develop dysuria with often mild, watery - slimy or mucous - purulent discharge. On examination the C. trachomatis urethritis only presents itself with a discrete redness of the orificium urethrae and an agglutination of the urethra. Clinically silent infections are common (up to 50%). If left untreated, the urethritis resolves itself within a few days or weeks. Exacerbations caused by infections, mechanical triggers (sports, vehement coitus, etc.) can be found. Initial infection and exacerbations are clinically indistinguishable. Complications such as clinically significant urethral strictures as well as asymptomatic urethral strictures (5% of all patients with healed C urethritis) can occur. (25-28,30-32)

- Epididymitis:

  By intracanalicular spread of the pathogen through the vas deferens an epididymitis can arise. This is usually one sided and in the acute stage manifests itself as a painful swelling and induration of the epididymis. Sometimes also the testicles are involved. In severe cases the pain radiates into the lower abdomen and groin. Furthermore the patients usually suffer from malaise and fever. As a late effect of bilateral infestation sterility can occur. (25-28,30-32)

1.2.3.2 Genital infection in women with C. trachomatis serovars D-K

Up to 80% of C. trachomatis caused infections in women are asymptomatic. The first and most common site of infection is the cervix. Thereafter, the infection can ascend to the endometrium, the adnexa, and the lesser pelvis as well as into the peritoneum. C. trachomatis may also lead to urethritis, which can occur either secondary or isolated. (25-28,32-35)

- Cervicitis:

  C. trachomatis only infects the high prismatic columnar of the endocervical canal and can lead to a mucopurulent cervicitis (MPC), which is charactarized by a purulent or mucopurulent discharge resulting in irritation, itching and burning of the introitus vaginae. Due to the low innervation of the cervical mucosa only 30% of infections cause discomfort. (25,26,28,32-35)

- Urethritis:

  In approximetly 60% of all cases of dys- and pollakisuria without bacteriuria among sexually active young women C. trachomatis is accountable. Only about half of the women infected have subjective symptoms such as dysuria or strangury. If symptomatic it is characterized by the lack of suprapubic tenderness, frequently preceded by new sexual relationships within the last months. (25,26,28,32-35)

- Pelvic inflammatory disease (PID):

  The term PID summarizes an inflammation of the organs and tissues of the female upper genital tract. These include endometritis, parametritis, salpingitis, oophoritis, tuboovarian abscesses and peritonitis. Mostly it is a polymicrobial infection caused by ascending spread of the pathogen through the cervix and into the uterus, the fallopian tubes and the peritoneal cavity. Often the causative organism cannot be determined. However, the most frequent causes include C. trachomatis and N. gonorrhoeae as well as aerobic and anaerobic organisms in the vaginal flora. Especially among young women in the Western world C. trachomatis and Neisseria gonorrhea (up to 75%, more than half of which C. trachomatis) are to be found most often. Typical symptoms of PID are lower abdominal pain, dyspareunia, fever, back pain and vomiting, and symptoms of infection of the lower genital tract such as discharge, bleeding, itching and unpleasant odor. The typical patient with PID is young (75% < 25 years) and shows rather mild discomfort although the course can range from asymptomatic subclinical manifestation to severe life-threatening illness. Possible damages include chronic pelvic pain, ectopic pregnancy and infertility. (25,26,28,32-36)

- Post-partum endometritis

  Delivery often triggers an exacerbation of a persistent chlamydial infection and can lead to infestation of the endometrium, often followed by fever. There is usually no enlargement or tenderness of the uterus. (25,26)

- Fitz-Hugh-Curtis Syndrome (Perihepatitis)

  Usually it is caused by direct ascension of a gonorrheal or chlamydial pelvic inflammatory disease or rarely metastatic mostly accompanied by acute pain of the right upper abdomen with tenderness and guarding. (25,26)

1.2.3.3 C. trachomatis serovars D-K infections in both sexes

- Adult chlamydial ophtalmia (AOC)

  The infection takes place through secondary spread of chlamydial infested genital secretions with an incubation period of one to two weeks. The infection presents itself as a mostly unilateral chronic follicular conjunctivitis with acute or subacute onset and mukois-mucopurulent secretion. After about a week, there is a growing follicular hypertrophy of the conjunctiva with a focus in the fornices. A typical late symptom is ptosis. Left untreated the AOC usually heals without permanent damage after months. (25,26)

- Sexually acquired reactive arthritis (SARA)

  SARA is a sterile synovitis based on immune mechanisms not yet completely clarified and an accessory symptom of urogenital chlamydial infections, as well as infection of the intestinal tract. (25,26)

- Proctitis

  In homosexual men up to 15% of proctitis is caused by Chlamydia trachomatis. In females it may be an expression of genital infection spreading into the rectum. The clinical presentation ranges from completely asymptomatic to light anorectal pain accompanied by itching and anal-eczema. Under proctoscopic examination, an erythematous, slightly vulnerable mucosa with mucopurulent secretion can be found. (5,25,26)

1.2.3.4 C. trachomatis serovars D-K infections in newborn children

- Inclusion conjunctivitis of the newborn (ICN)

  ICN occurs in approximately 30% of newborns of infected mothers, starting 5-14 days after birth with a mucopurulent conjunctivitis. After two to three weeks it usually heals spontaneously without permanent damage. The Credé-prophylaxis can not prevent the formation of chlamydial conjunctivitis in the newborn. (25,26)

- Rhinitis

  Symptoms range from mild to severe nasopharyngeal obstruction and sometimes are considered to be a typical prodrome of Chlamydia associated pneumonia. (25,26)

- Pneumonia/ Otitis media

  Approximately 3% of newborns of infected mothers develop a bronchitis/pneumonia and/or otitis media. Incubation period of this mostly afebrile pneumonia ranges from two weeks to four months with a peak after ten weeks. These newborns show a hacking, dry cough and inspiratory stridor. The course is usually self-limiting but obstructive pulmonary changes may be a possible long-term consequence. (25,26)

1.2.4 Diagnostics

All forms of Chlamydia infection can only be diagnosed by the detection of the pathogen. In order to proof a chlamydial infection the direct pathogen detection test is the method of choice. In general the selection of the most appropriate method depends on the clinical appearance and the disease suspected most likely. Using the appropriate test material is crucial for diagnosis. Not only inflammatory exudate, but also epithelial cells are required and converted into the corresponding transport medium to the laboratory. Taking of urethral respectively cervical swabs is the standard procedure to gain specimen for an oculogenital infection caused by Chlamydia trachomatis. Additionally modern amplification methods (PCR, LCR) are applied in order to detect antigens of pathogens using centrifuged fresh micturition (ten to thirty ml). (25-27,37)

Inadequate technique while collecting a sample is a common source for a fault diagnosis of chlamydia. To minimize the risk of a false positive or negative result the correct usage of a swab has to be trained. In males the swab gets introduced two to four cm into the urethra and must be rotated at least 15 times with light pressure against the urethral wall to gain enough epithelial cells for a specimen. In women the smear is taken from the cervical

canal. First, the cervix gets cleaned of preexisting mucus and exudate. After that the swab gets inserted into the cervical canal and is rotated as mentioned above. To avoid a false positive result of the test and to prevent contamination with bacteria from the vaginal wall the swab should be withdrawn carefully. Similar techniques are used to get smear of the rectal mucosa favorably by means of a proctoscope. (25-27,37)
Chlamydia trachomatis can be proven by the following methods:

- Isolation in cell culture

  Until the introduction of the antigen detection and biomolecular methods the cultivating of Chlamydia trachomatis in cell cultures (mostly McCoy - cells) was the easiest way of proofing an infection. Its implementation is associated with considerable technical effort but due to high sensitivity (80%) and specificity (approximately 100%) it still is "gold standard" for detecting Chlamydia. Positive culture results are always a sure indication of a chlamydial infection. Negative results are approximately in 20% false negative. However high costs and time efforts (48h), transport to special laboratories and non-uniform standardized culture methods are disadvantages. (25-27,37)

- Antigen detection

  This method is trying to examine and detect antigens of the pathogen. For this purpose either the species-specific MOMP antigen for C. trachomatis or the genus specific lipopolysaccharide antigen can be used. The DIF (direct immunofluorescence assay/ MOMP antigen) is the simplest and cheapest method with high specificity and sensitivity (both 90%). Fluorescein-labeled species-specific antibodies are used to stain the extracellular elementary bodies. The ELISA (enzyme linked immunosorbent assay) detection (lipopolysaccharide antigen) is suitable for quick and objective testing of large sample sizes, but has to be confirmed by other tests (DIF), because of its possibility of cross-reactions with other bacteria and subsequent false-positive tests. (25-27,37)

- Nucleic acid hybridization (NAH)/ Nucleic acid amplification (NAA)

  The nucleic acid hybridization method is based on the detection of ribosomal RNA of Chlamydia trachomatis, using a single-stranded DNA probe in the chemiluminescence process. Today NAA's are able to detect Chlamydia trachomatis with a very high

sensitivity (>90%) and specificity (>99%) and make older methods obsolete in most cases. Commercially available test systems for C. trachomatis with amplification include the polymerase chain reaction (PCR) - test Amplicor©, the ligase chain reaction (LCR) - test LCX© and the transcription mediated amplification (TMA) assay. The key advantage of DNA amplification methods is the possibility of using swabs from the cervix or urethra as well as small amounts of first-void urine or a vulvovaginal-smear with neclectable loss of sensitivity. The DNA evidence is done in three steps. DNA gets extracted from the test sample and during preparation included enzymes are inactivated. The second step is carried out by a thermocycler, in which the sample DNA sequence is amplified and thereby increased exponentially. At last the multiplied DNA gets detected by hybridization (PCR) or ELISA (LCR). (25-27,37-39)

- Serology
  Genus-specific tests (r-ELISA) detect antibodies against all chlamydial species. Species-specific tests (peptide ELISA), using recombinant peptides of Chlamydiae, identify single specific antibodies against different chlamydiae species. The first antibodies after an infection with C. trachomatis are usually measured traceably after six to eight weeks, and therefore are not necessarily to be expected at an acute infection. Following an infection with C. trachomatis the antibodies can persist for months or years, so that often it cannot be distinguished between past and current infections. Positive serological findings in the absence of symptoms should therefore not be interpreted as necessarily persistent infections. Summarizing the determination of Chlamydia antibodies in uncomplicated urogenital infections is of no significance. However in ascending urogenital infections, high anti-chlamydial IgG and IgA antibody titers can confirm the suspected diagnosis. (25-27,37)

1.2.5 Therapy

To understand the therapeutic approaches one must first examine the chlamydial biology. For example, the envelope of the gram-negative pathogen does not express any peptidoglycan, although genome analyzes have shown that the cells have an almost complete pathway for peptidoglycan including penicillin-binding proteins. However, the peptidoglycane genes as well as the genes responsible for nucleoside triphosphate (ATP) seem to be shut off. This being the reason that β-lactam antibiotics such as penicillin are

ineffective due to the lack of penicillin-binding proteins in the cell wall. Chlamydiae are susceptible to a range of antibiotics that interfere in the DNA and protein synthesis, including tetracyclines and macrolides. Because of the intracellular localization of the pathogens all antibacterial agents must be able to penetrate the host cell wall according to achieve an adequate intracellular concentration.

Genital chlamydial infections should be identified and treated as early as possible. (40) In the past, given the long life cycle of C. trachomatis (48 -72 hours) a multiple treatment was used. For decades, the daily dose of doxycycline 100 mg 2 x 1 or erythromycin 500 mg 4 x 1 for seven days was considered standard. Alternatively a treatment with ofloxacin 2 x 300 mg or levofloxacin 500 mg 1 x 1 each for seven days were additional treatment options. The introduction of azithromycin with its long half-life in tissue now allows a single dose treatment of genital chlamydial infections. (41) A meta-analysis of twelve clinical studies in which the therapeutic efficacy of a single dose of azithromycin was compared with a seven-day doxycycline therapy showed a nearly 100% microbiological eradication in both treatment regimens. Both drugs were equally tolerable. (42) The U.S. Centers for Disease Control and Prevention (CDC) currently recommends a single dose of 1g of azithromycin or a 7-day doxycyclin treatment as "first-line regimen" for the treatment of uncomplicated genital C. trachomatis infections among teens and adults of both sexes. (43) Studies with erythromycin have proved less efficient and showed gastrointestinal side effects in patients. After completing a successful treatment, a re-examination after three months is recommended considering the high re-infection risk particularly among young women. (43,44) Relapses, despite the antibiotic therapy, occur. For that reason treatment with doxycycline or erythromycin in uncomplicated genital infections should be continued for fourteen days. Timely treatment of genital C. trachomatis infection does not only prevent the further spread of the pathogen but also protects patients against complications especially pelvic inflammatory disease (PID) and tubal factor infertility. In complicated infections such as pelvic inflammatory disease or epididymitis a two weeks, possibly parenteral, therapy is recommended. Sexual contacts may only take place after completion of treatment. Because of the risk of infection and the possibility of a false negative result all sexual partners within the last 60 days should be co-treated. (43,45,46)

A specific treatment regimen is required in C. trachomatis-infected pregnant women. Doxycycline and quinolones are contraindicated in pregnancy. A single dose one gram of azithromycin was found to be safe and effective as well. (47,48) The CDC also recommends amoxicillin as "first-line treatment regimen". Amoxicillin was found to be

more effective and better tolerated than erythromycin, which was used for long years. (43) Pregnant women, who have been treated during the first trimester, should be re-examined three to four weeks after completion of therapy, followed by a further check up three months later. (43,47,48)

1.3 Neisseria gonorrhoeae

1.3.1 Etiopathogenesis

Gonorrhea is known since ancient times; however it was not clearly distinguished from syphilis. Like many other sexually transmitted diseases gonorrhea was interpreted as a variant of syphilis in those days. In 1767, John Hunter, a Scottish surgeon conducted a study in a self-attempt to separate syphilis from gonorrhea. He inoculated himself with gonorrheae, using a needle that was unknowingly contaminated with syphilis. Experiencing both symptoms of syphilis and gonorrhea he therefore concluded that both diseases were transmitted by the same pathogen and published the results in his 1786 written book "A Treatise on the Venereal Disease". (49) Only in 1867, Ricord could indicate that there were two separate diseases. The dermatologist Albert Neisser then provided the final proof. In 1879 he identified Neisseria gonorrhoeae or the Gonococcus. In 1882, it was Leistnikov who was the first to culture the pathogen and in 1906 the first serological gonorrhea test was introduced. (50)
With about 88 million infections annually, gonorrhoea is the fourth most common STI worldwide and the second most common bacterial STI in Europe. (51) In Western countries, the incidence of gonorrhoea reached a peak by the end of the Second World War. Since 1980, gonorrhea subsides in western countries, although its occurrence in some countries is underestimated because of infrequent case reporting and surveillance. Due to this fact the distribution of gonorrhoea is subject to a wide spectrum of geographical variation. Men are usually statsitically overrepresented by reason of fewer asymptomatic cases. Women are most common affected between the ages of 15 to 19, men between 20 to 29 years. (51-53)
N. gonorrhoeae is a human pathogenic bacteria that cannot cause a natural infection in animals and therefore is restricted to a limited human reservoir. The transmission from human to human, usually takes place through sexual contact. Gonococci are susceptible to desiccation and therefore would not survive outside of their host. Asymptomatic infected persons are the most important source and reservoir of infection. Gonorrheae can also be

transmitted during birth from an infected mother to her child (vertical) and in newborns mostly manifests itself as eye infection (ophtalmia neonatorum). (54-56)
For gonococci the vagina and the penis are the usual portals of entry into the body, but because of certain sexual practices, they can also get into throat or anus. Gonococci prefer the cylindrical ephitelium of the female and male urethra, the cervix, rectum, and the conjunctiva. While the squamous epithelium lined vagina of adult woman is spared from the agents, the higher vaginal epithelium of prepubertal girls can get easily infected. (54-56)
N. gonorrhoeae attaches itself to host cells, using adhesins on its surface and misuses other membrane components to damage the epithelial cells. After their adhesion, the gonococcus begins to multiply rapidly and ascends via cervix and urethra. N. gonorrhoeae does not produce any exotoxin. The damage to the host tissue is based solely on inflammatory reactions that are caused by the gonococcus. A sustained, untreated infection can lead to chronic inflammation and fibrosis. Usually, the infection is limited to the genital tract, but in some cases bacteria are isolated, which have spread via bloodstream to other parts of the body. The clinical and the microscopic image are shaped by pus. Gonorrhea does not leave any immunity. Ascending infections can lead to sterility, hematogenous spread to disseminated gonococcal infection. Gonorrhea is still one of the most common sexually transmitted diseases worldwide, and thus an important indicator of the effectiveness of preventions for HIV and STD in general. (54-56)

1.3.2 Path of infection

Transmission is almost exclusively through sexual intercourse. Non-sexual transmission via smear infection or contact with contaminated objects (toilet articles, thermometer) is usually only possible in girls before puberty, who are not yet protected by acidic vaginal pH (gonorrheal vulvovaginitis infantum). Child abuse must therefore be excluded! Intra partum gonorrhea can be transmitted to the newborn, leading to gonoblennorrhoe, formerly the main cause of childhood blindness. Since the introduction of Crede - prophylaxis this complication is almost gone. (54-57) The likelihood of an infection after contact with an infected partner depends on the number of germs and also on the gender: women who have sex with an infected partner almost always get infected (80%), men, however, only at a rate of about 20%. Gonorrhea can also be acquired through anal and oral sex. (54,55)

1.3.3 Clinical Pathology

In the first two to seven days of infection patients can develop typical symptoms such as urethral discharge and pain during urination in men and discharge from the vagina in women. In asymptomatic or atypical courses it is often difficult to determine the time of infection. At least 50% of infected women have mild or no discomfort at all and the infection only expresses as imperceptibly increased discharge. This being the reason that they are not consulting a doctor, they remain untreated and continue to be contagious for others. Under some circumstances women will stay unaware of the infection until complications such as pelvic inflammytory disease (PID), chronic abdominal pain or infertility caused by tubal occlusion appear. (54,55,58,59)

In the acute phase, the symptoms of N. gonorrhoeae in infected men are usually stronger and rarely go unnoticed. The infection begins in the fossa navivcularis or cervix and spreads ascendant. Gonococci are epithelial parasites with a predilection for columnar epithelium (in man the urethra and its appendages, prostate, and epididymis, in women the Bartholin's glands, urethra, cervix and tubes). Gonococcal infection of the throat can lead to a sore throat and an infection of the rectal mucosa may result in purulent discharge.

The urinary bladder, upper urinary tract, vulva, vagina and endometrium are hardly ever affected. If left untreated, the acute infection devolves into a subacute or chronic phase with fewer symptoms that can lead into a spontaneous healing after six to twelve months.

A protective immunity against re-infection does not occur. (54,55,58,59)

1.3.3.1 Association with urogenital chlamydial infection

Chlamydia trachomatis D - K causes ascending infections of the urogenital tract, which in many aspects are similar to gonorrhea, but milder in distinct. It is often transmitted together with gonorrhea (50% of cases of gonorrhea). Because of its long incubation period and resistance to gonorrhea standard treatment, C. trachomatis appears like a resulting sickness of gonorrhea. It is interpreted quite often as a relapse and can cause confusion because of an alleged resistance of the gonococcus to penicillin. Therefore any case of gonorrhea should also be checked for chlamydia trachomatis. (54,55,60)

1.3.3.2 Gonorrhea in men

- Acute gonococcal urethritis:

  Acute gonorrhea in men starts with an infection limited to the pars spongiosa uerthrae, refered to as urethritis anterior. First symptoms are a tingling and burning sensation of the fossa navicularis of the urethra, which is reinforced during urination (dysuria). Usually within 24 hours it is followed by a purulent, creamy, yellow to greenish-yellow and abundant urethral discharge. Within two to six days after infection the majority of men develop a massive urethral discharge and dysuria. (It is said that military doctors often used to diagnose gonorrhea in the field just by looking at the underpants.) The orificium urethrea may also be swollen and red. About 25% of all patients never develop more than a scanty serous discharge, which is indistinguishable from a Chlamydia or NGNCU based discharge. These men will have no symptoms during the day. Only in the mornings before the first micturition, a drop of pus will be expressed. In about 10% of infected men, gonorrhea takes a completely asymptomatic course. If the infection ascends across the external urethral sphincter without treatment for 10-14 days a urethritis posterior develops. This can be accelerated by mechanical influences. The outcome is an enhancement of dysuria, urgency, and in severe cases a terminal haematuria. Simultaneously patients suffer from fever, malaise and joint pain. As a complication of the urethritis posterior an infection of the Cowper's glands up to a Cowper's abscess can occur, located as a cherry-sized redness and swelling behind the scrotum. A Cowperitis may lead to a narrowing of the posterior urethra (Cowper's stricture). (54,55,59)

  The ascending gonorrhea infection may also lead to prostatitis, vesiculitis, funiculitis and epididymitis. In 40% of homosexual men an isolated rectal gonococcal infection can be observed by direct inoculation of the rectal mucosa during anal sex. Diseases of the individual sections of the male sex organs are clinically almost indistinguishable from each other. Urethral discharge may be completely absent in ascending gonorrhea, although the Gram stain urethral swabs are often positive. Except Neisseria gonorrhoeae, also Escherichia coli, Streptococcus faecalis, Staphylococcus aureus and Chlamydia trachomatis can cause ascending infections. (54-56,59)

- Chronical gonococcal urethritis:

  Untreated, the acuity of gonorrhea infection keeps declining gradually. The amount of discharge decreases and its texture becomes slimier. After weeks to months, gonorrhea

becomes asymptomatic. Residual symptom is the so-called bonjour-drop (during the night accumulated mucus droplets in the urethral orifice). This development might lead to spontaneous healing, but usually it results in a mild chronic gonococcal urethritis, which by periodic exacerbations (sex, cold, comorbidities) can lead to complications (strictures, prostatitis, epididymitis). (54-56,59)

1.3.3.3 Gonorrhea in women

- Acute gonorrhea:

  In women, gonorrhea generally proceeds mild and uncharacteristic in the initial phase. In about 50% of all cases it remains unnoticed at all. Therefore, the risk for development of chronic gonorrhea is higher in women. Severe systemic complications are more common, and infectivity is considerably longer. Gonorrhea in women begins, unlike in men, often at multiple locations at the same time: (54-56,58)

  - Urethritis (75%)
  - Cervicitis (90%)
  - Proctitis (40%)

  The cervix with the cervical canal is the most common site of infection. In 70-90% of women with cervicitis gonorrhoica a concomitant urethritis is present. Increased vaginal discharge is the guiding symptom. Subjectively sharp pain or burning sensation exist during micturition, which can increase into strangury based on bacterial cystitits. Menorrhagia and metrorrhagia are symptoms of the co-participation of the endometrium. Around 50% of women with urogenital gonorrhea have no complaints, a crucial factor in the epidemiology of gonorrhea proliferation. (54-56,58)

  During most clinical examinations a yellow to yellowish-white discharge from the cervical canal can be found, as well as redness and swelling of the vaginal portion and contact bleeding. Pus from the urethra can be expressed. (54-56,58)

  Local complications of an infection with gonorrhea can be an infestation of the urethral glands (Skene's glands) and the vestibule glands (Bartholin's glands). The retention of exudate can create a Bartholin's abscess. Ascending gonorrhea in women conducts via the endometrium and the tubes to the ovaries and further into the pelvic-peritoneal region, where often a separation of individual diseases is not possible anymore.

Clinically, the majority distributes into cases of salpingitis, adnexitis or an infection of the entire pelvis (pelvic inflammatory disease, PID). If the gonorrheal infection ascends even further from the tubes via the abdominal cavity into the perihepatic area, it can cause an acute perihepatitis. (54-56,58)

These complications appear in almost one third of the cases. Common initial symptoms reach from acute spasmodic, usually unilateral pain in the lower abdomen after menstruation, fever up to 39 ° C, to general symptoms and leukocytosis. The clinical diagnosis of gonorrhea in women is difficult because of the same or similar symptoms caused by C. trachomatis, T. vaginalis, C. albicans, H. simplex virus and a number of other microorganisms. (54-56,58)

- Chronic gonorrhea:

  Untreated acute gonorrhea develops into oligosymptomatic states as, mucopurulent discharge, recurrent portio erosions, retention cysts of the cervix, mild chronic urethritis, recurrent bartholinitis and bartholin cyst. The diagnosis is difficult because smear and culture often remain negative. Final state is most often a chronic adnexitis. Permanent disabilities result from scarring fibrosis: tubal occlusion, infertility, high risk of tubal pregnancy, adhesions of the uterus, hydrosalpinx, tubo-ovarian cysts. (54-56,58)

- Gonorrhea in pregnant women:

  The significance of gonorrhea in pregnant woman is because of its associated complications such as premature rupture of membranes, preterm labor, chorioamnionitis and septic abortion. Without treatment the newborn is endangered to contract an oropharyngeal infection or ophthalmia neonatorum. These consequences of a gonococcal infection justify a routine monitoring at the first pregnancy examination. Pregnant women from STI risk groups should receive a second check up during the 36th to 38th gestation week. (54,58,61,62)

1.3.3.4 Gonorrhea in children

Unlike the vagina of sexually mature woman the cylindrical epithelium of the vestibule and the vagina of pre-pubertal girls offer favorable conditions for a gonococcal infection. In children all other parts of the urinary and genital apparatus may be involved in the

infection as well. The symptoms of vulvo-vaginitis gonorrhoica infantum range from itching, purulent discharge up to strong dysuria, loss of appetite, constipation, and insomnia of the child, as well as asymptomatic. In all cases of genital diseases with purulent discharge in children gonorrhea has to be considered. Infections through contaminated linen, towels, sponges, toilet seats, thermometers, or toys are not proven. Therefore, when detecting gonorrhea in children sexual abuse should be excluded. (54,55,63)

1.3.3.5 Extragenital manifestations of gonorrhea

- Rectal gonorrhea:

  An infection of the rectal mucosa can be detected in about half of the women with an urogenital gonococcal infection, mostly due to secondary contamination from the genital secretions. In homosexual men, the rectum is a common primary site of infection. The rectum as sole location of infection is found in about 5% of women with gonorrhea and 40% of homosexual men. In women, the rectal gonorrhea often runs asymptomatic, while about two thirds of the men express complaints. Symptoms range from anal pruritus, haemorrhoids to a classic proctitis with rectal pain, tenesmus, bloating and constipation, occasionally presenting with perianal redness and discharge. During proctoscopic analysis redness, slight vulnerability and purulent deposits on the mucous membrane are to be found quite often. The removal of material for microscopic examination and culture should be done under proctoscopic view as well. The asymptomatic rectal gonorrhea represents a reservoir of infection for gay men. (54-56,64,65)

- Pharyngeal gonorrhea:

  In only about 5% of the cases the throat of gonorrhea infected patients is the sole side of infection. 8-22% of women, 3-7% of the heterosexual men and 11-25% of homosexual men suffering from urogenital gonorrhea, also have an involvement of the pharynx. Transmission occurs through orogenital contact. In approximately 90% of the cases infection runs asymptomatic, this being the reason to routinely take pharyngeal swabs of STI patients from the back of the throat wall and tonsils. Because of the occurrence of asymptomatic meningococcal and some non-pathogenic Neisseria species the confirmation of a pharyngeal gonorrhea diagnosis can only be done by culturing the pathogen. (54-56,64,65)

- Ophthalmoblennorrhoea neonatorum:

  Gonococcal conjunctivitis of the newborn can be caused by intrauterine infection, or infection during delivery. The risk of intrapartum infection from an infected mother is around 30-50%. Symptoms of an acute purulent conjunctivitis occur within five days after birth. The children usually develop a crude swelling of the eyelids, followed by increasing purulent secretion in both eyes. The eyes are extremely painful and sensitive to pressure. Without treatment, the disease can quickly spread to the cornea and lead due to ulceration and perforation, to secondary glaucoma and blindness. (54-56,64)

  In the 19th century more than half of all blind people lost their sight because of gonococcal ophthalmia. Today, the incidence of exposed children, who received the in 1881 by Karl Credé invented silver nitrate prophylaxis (Credé prophylaxis), lies between 0 to 5%. In exposed children who did not receive prophylaxis, 2 to 30% are affected. In addition to the 1% silver nitrate solution, a 0.5% erythromycin or a 1% tetracycline eye ointment can be used as a single application for prophylaxis. A simultaneous oropharyngeal gonococcal infection occurs in 35% of children suffering from ophthalmia. A gonococcal infection must be excluded in any conjunctivitis of the newborn. (54-57,64)

- Ophthalmoblennorhoea adultorum:

  Gonococcal ophthalmia in adults generally runs stormy and has a worse prognosis than in newborns. Usually one eye will be infected in patients with concurrent anogenital gonorrhea. Transmission occurs by autoinoculation. Patients with anogenital gonorrhea should be apprised of the potential for transmission to the eyes. The symptoms consist of watery eyes, light-shyness, burning and itching. (54-56,64)

- Disseminated gonococcal infection (DGI):

  DGI occurs in about 0.5-3% of the patients with a local gonorrhea. It is much more common in women (60-97%) than in men. In female patients, the first symptoms usually occur within eight days after the last menstruation, after delivery or abortion. DGI is clinically characterized by the triad of fever, acute arthritis and vasculitic dermatological reaction. It usually starts with arthralgia and tendosynovialitis. A significant arthritis with joint effusion occurs in 30-40% of patients in the course of the

disease. Fever, leukocytosis, or skin manifestations occur in 50-75% of patients. Overall, a DGI is rarely thought of, because of the often asymptomatic local gonococcal infection and the uncharacteristic systemic disease. Evidence of systemic gonococcal infection is difficult. Bacteremia progresses in waves, and with the course of the disease, the blood cultures rarely succeed. Only out of 20-30% of all DGI patients, gonococci of the blood can be cultivated, and less than half out of synovial or skin biopsies. (54-56,64,66)

1.3.4 Diagnostics

Basically, the direct detection of pathogens, culture, antigen detection, DNA hybridization and amplification methods are available for disposal. The direct detection of the pathogen has the significance of a most adequate screening examination, but has no probative value. Only cultural identification has probative value. Antigen and DNA detection methods are particularly useful if transportation problems affect the quality of the culture. Serological methods are practically meaningless. (54,55,64,67-70)

The diagnosis of gonorrhea is made by detection of the pathogen in the microscopic preparation and culture. In women, an endocervical and urethral swab is taken. By using additional anal and pharyngeal swabs, the detection rate can be increased. In men, an urethral swab is taken. Before taking the urethral sample the patient should not have urinated for at least four hours. Preferably the examination takes place in the morning before the first micturition. In homosexual orientated patients also an anal and a pharyngeal swab must be taken. In the symptomatic urethritis of men, the sensitivity of the Gram stain is 95%. In asymptomatic men the sensitivity decreases to 40-70%. In presence of clinical symptomatic the specificity of a typical Gram staining achieves almost 100%. If confronted with a suspicious case history without clinical symptoms, unexplained symptoms or a doubtable gram stain a cultural test is necessary. A culture is also essential for determining the antibiotic sensitivity of the pathogen and forensic questions. The cultural isolation and identification of Neisseria gonorrhoeae is still the diagnostic standard for the detection of gonococcal infection. Gonococci do not tolerate dryness and should immediately be inoculated on a culture medium after taking swabs. After the identification of Neisseria gonorrhea a ß-lactamase test should be performed to determine the plasmid-mediated penicillin resistance. (54,55,67,69,71,72)

1.3.5 Therapy

The resistance profile of N. gonorrhoeae has evolved over the decades. Nowadays, the percentage of ß-lactamase generating N. gonorrhoeae strains lies at 25% worldwide. In the U.S. it reaches up to 30%, in Asia and in Africa up to 90%, and in Germany up to 3-6%. Gonococcal infections with chromosomal spectinomycin and plasmid-mediated tetracycline resistance have been described. Lately the occurrence of gonorrhea infections by means of ciprofloxacin-resistant strains has increased worldwide. Still drug of choice today is ceftriaxone. High serum concentrations of the antibiotic are usually sufficient because of the short generation time of the gonococci. Due to the often accompanying C. trachomatis infection (10-30%) a combination with azithromycin or doxycycline is recommended. (54,64,67,73)

Infection	Recommended regimen	Alternative regimens
Uncomplicated gonococcal infections of the cervix, urethra, and rectum	*Ceftriaxone 250 mg* in a single intramuscular dose plus *Azithromycin 1 g* orally in a single dose or *Doxycycline 100 mg* orally twice daily for 7 days	*Cefixime 400 mg* in a single oral dose plus *Azithromycin 1 g* orally in a single dose or *Doxycycline 100 mg* orally twice daily for 7 days plus Test-of-cure in 1 week If the patient has severe cephalosporin allergy: *Azithromycin 2 g* in a single oral dose plus Test-of-cure in 1 week
Uncomplicated gonococcal infections of the pharynx	*Ceftriaxone 250 mg* in a single intramuscular dose plus *Azithromycin 1 g* orally in a single dose or *Doxycycline 100 mg* orally twice daily for 7 days	

Disseminated Gonococcal Infection (DGI)	*Ceftriaxone 1 g* IM or IV every 24 hours	*Cefotaxime 1 g* IV every 8 hours or *Ceftizoxime 1 g* IV every 8 hours
Gonococcal Infections Among Children	• Children weighing more than 45 kg get the same treatment as recommended for adults • Standard treatment for Children weighing 45 kg or less with uncomplicated gonococcal vulvovaginitis, cervicitis, urethritis, pharyngitis, or proctitis: *Ceftriaxone 125 mg* IM in a single dose • Standard treatment for Children weighing 45 kg or less with bacteremia or arthritis: *Ceftriaxone 50 mg/kg* (maximum dose: 1 g) IM or IV in a single dose daily for 7 days	
Ophthalmia Neonatorum Prophylaxis	*Erythromycin* (0.5%) ophthalmic ointment in each eye in a single application	*Ceftriaxone 25-50 mg/kg* IV or IM, not to exceed 125 mg in a single dose
Pregnancy	*Azithromycin 2 g* orally	

Table 1 – Treatment guidelines for N. gonorrhoeae (74)

Follow-Up:

Not the subjective condition of the patient, but only the negative bacterial inspection results are crucial for the healing of gonorrhea after the therapy. Three to seven days after completion of therapy, swab samples from all previously infected sites should be taken for control. If necessary several check-ups can be performed, for women at best immediately after the next menstrual period. If symptoms of urethral infection are persistent or recurrent after treatment, reasons may be a treatment failure, reinfection, an additional chlamydial urethritis or a non-gonococcal, non-chlamydial urethritis (NGNCU).

The pathogen detection and identification should be repeated from all infected sites. Because of the extended length of the incubation period of a Chlamydia induced urethritis, swabs should be taken at the latest at the first gonorrhea check-up. All potential sexual partners of patients should be examined and if necessary treated. Examination and treatment of partners is not only important for epidemiological reasons, but also serves to protect the sexual partners from disease and the patients themselves against re-infection. (54,55,75-77)

1.4 Mycoplasmataceae

1.4.1 Etiopathogenesis

The Mycoplasmataceae family, part of the Mollicutes class, includes the two genera Mycoplasma and Ureaplasma, which usually colonize the urogenital tract of humans asymptomatically. In 1898, for the first time ever, mycoplasma was isolated by Nocard and Roux in cattle and in 1937 Dienes and Edsall detected and isolated mycoplasma in humans. (78)

Mycoplasma and Ureaplasma are the smallest freeliving, self-replicable microorganisms in terms of genome size and cellular dimensions that can exist outside a host cell. They are characterized as immobile, pleomorphic, gram-negative bacteria and belong to the mollicutes, a class of bacteria without rigid cell wall structures and therefore high morphological flexibility that allows them to present themselfes in mushroom-like forms. They can also penetrate through bacteria-proof filter due to its flexible form. They differ from other bacteria by their small cell size and the small genome, which is lacking the genes coding for the cell wall and contain cholesterol. Otherwise cholesterol is only found in eukaryotic cells, making conventional techniques for light microscopy such as Gram staining inapplicable. (79,80)

The high demands of Mycoplasma and Ureplasma on culture media as well as their long generation time and slow growth over several days make the cultural pathogen detection complex and timeconsuming. Futhermore, antibiotics that interfere with cell wall synthesis, such as ß – lactam antibiotics, are clinically irrelevant. Both Mycoplasma and Ureaplasma are very susceptible to dehydration and therefore specialize in parasitic colonization of mucous membranes of the respiratory and urogenital tract. When adhering to objects, mycoplasma and ureaplasma survive only a very short time and are inactivated rapidly by disinfection processes. (79,80)

Of the seven Mycoplasma-species (M. hominis, M. genitalium, M. penetrans, M. primatum, M. spermatophilum, M. fermentans, M. pneumoniae) and two Ureaplasma – species (U. urealyticum, U. parvum) which can be detected in the mucous membranes of the urogenital tract, only M. hominis and U. urealyticum have been examined in the retrospective study. (81)

U. urealyticum differs from other mycoplasma by forming very small colonies (T-Mycoplasma). Another important biochemical distinguishing criterion between mycoplasma and ureaplasma is the urea - test, which is only positive for ureaplasma, because of the ability to degrade urea. In older literature U. urealyticum was divided into biovars 1 and 2, but due to the amplification method, we now distinguish U. urealyticum from U. parvum. There was doubt about the sole pathogenicity of U. urealyticum, because it often could be found accompanying a N. gonnorhoeae or C. trachomatis induced urethritis, but there are reasons for its at least potential pathogenicity: (79-82)

- U. urealyticum is sometimes grown as a sole pathogen at non- gonorrheal- non-chlamydial- urethritis (NGNCU).
- After treatment with appropriate antibiotics the NGNCU heals and U. urealyticum cannot be detected anymore.
- When using an in vitro ineffective antibiotic against U. urealyticum the NGNCU as well as the pathogen persist. (80)

1.4.2 Path of infection

Genital mycoplasma and ureaplasma are facultative pathogens, which often colonize the genital tract asymptomatically. Their importance for the development of sexually transmitted diseases is controversial. Mycoplasmatacaeae are predominantly sexually transmitted and also can be passed on from the mother to the newborn during birth. Colonization of the genital tract with mycoplasmataceae increases with the number of sexual partners, so that U. urealyticum in particular must be considered as part of the genital flora in sexually active young people. The incidence of detecting mycoplasma and ureaplasma positively correlates with young age and high sexual activity with variing partners. (78-80)

1.4.3 Clinical Pathology of Mycoplasma hominis and Ureaplasma urealyticum

Symptomatic infections with U. urealyticum are more likely in men. M. hominis rather colonizes the epithelial cells of the urogenital tract in women, but both species may also be found together in one host. Mycoplasma infections can cause clinical symptoms next to mute gradients. A possible explanation for this inconstant behavior is that only some serotypes are pathogenic, and that a minimum number of germs are required for showing

clinical symptoms. U. urealyticum may cause an infection (NGNCU) the first time the urethra is affected, but following contacts result in colonisation without disease, this being the reason for frequent isolation of ureaplasma from the urethras of healthy men.
Table 2 pictures the possible diseases and their likelihood related to an infection with U. urealyticum or M. hominis. An initial infection with U. urealyticum is similar to the gonorrheal anterior urethritis and expresses itself through a whitish serous discharge, associated with itching or burning in the urethra and dysuria. The diagnosis of NGNCU can only be made after exclusion of an infection with Neisseria gonorrhoeae and Chlamydia trachomatis. NGNCU caused by genital mycoplasmas display a high rate of treatment failures and chronic recurrent courses if the pathogen cannot be detected successfully. (79-81)
The role of M. hominis and U. urealyticum in prostatitis, epididymitis, SARA as well as in infertility is still little studied and not yet determined.

Disease	Ureaplasma urealyticum	Mycoplasma hominis
NGNCU	+++	-
Cervicitis	-	+++
PID	-	+++
Bacterial vaginosis	+	+++
Prostatitis	?	?
Epididymitis	?	?
SARA	?	-

Table 2 – Association of infectious diseases with U. urealyticum/ M. hominis (81)

1.4.4 Diagnostics

Because of size and low affinity to dyes, the detection of mycoplasmas is not possible in Gram preparations. The best material for the detection of mycoplasma is the urethral swab, morning urine, as well as in women vaginal and cervical swabs. U. urealyticum and M. hominis can be detected culturally. With a calibrated loop, which allows a quantitative study, urethral secretion is placed in a special transport medium, transported at 4 ° C within 24 hours to a laboratory and inoculated on special culture media (mycoplasma agar). U. urealyticum grows within a week in the form of colonies, which because of their size (15-60 microns in diameter) are only visible microscopically. (79,80)

M. hominis colonies are larger (200-300 microns in diameter) and typically have a fried egg shape. Serological investigations for the diagnosis of infections with Mycoplasma are clinically irrelevant.

1.4.5 Therapy

The indication for therapy is not strictly bound to the detection of the pathogen, but should be considered at the presence of clinical symptoms and the absence of other causes. Since mycoplasmas have no cell walls they are resistant to ß-lactam antibiotics and cephalosporins. The agents of choice for Ureaplasma infections are tetracycline, macrolide antibiotics and quinolones. Doxycycline, 100 mg twice daily for 7 - 14 days is the standard treatment. If tetracycline cannot be used, as an alternative regimen, erythromycin is available in a dosage of 500 mg four times daily for 7-14 days (or in chronic urethritis up to 6 weeks). Also, ofloxacin, 400 mg twice daily for 7 days is effective. (74,79) If the urethritis is persistent after doxycycline treatment the cause might be doxycycline-resistant U. urealyticum. In that case an alternative regimen of metronidazole 2g orally in a single dose or tinidazole 2g orally in a single dose would be advisable. (74) For the treatment of M. hominis infections Clindamycin (300 mg orally twice daily for 7 days) is recommended. (83) Difficulties in treatment are often due to lack of adherence to treatment, re-infection and lack of partner evaluation, as well as insufficient clarification of the cause or ignoring the resistance situation.

1.5 Trichomonas vaginalis

1.5.1 Etiopathogenesis

T. vaginalis is a protozoan parasite that causes an infection called Trichomoniasis. It was discovered in 1836 and is a worldwide occurring STI, which is not notifiable in Austria.
It is considered to be the most common curable STD. For example in the United States, an estimated 3.7 million people have the infection, but only about 30% develop any symptoms of trichomoniasis. The WHO speaks of 248 million infections annually worldwide. (51,84,85)
T. vaginalis is a flagellate of oval shape with four flagella and an undulating membrane with 4-45 μm in length and 2-14 μm in width. It is clearly visible under the microscope, due to its characteristic convulsive movements. T. vaginalis preferably infects keratinizing mucosal surfaces, like the genitourinary tract and usually causes harmless, subjectively however, sometimes intense symptoms. (86,87)

There are significant differences in the infection rate between different population groups and between developed and developing countries. The peak age corresponds to that of the highest sexual activity and correlates with the occurrence of other STI's (gonorrhea, chlamydial infection). The exact prevalence of T. vaginalis is not collectable however women are affected more often and have symptoms more often than men. Depending on the study population and the method used for the diagnosis, the incidence of trichomoniasis in symptomatic female patients varies from 5% to more than 50% and in symptomatic men from 1% to 20%. According to some studies, 20% of women between the ages of 16 to 35 years suffer from a T. vaginalis infection at least once. Among gynecological patients about 10-20% and among prostitutes 50-70% is infected. About 70% of women with gonorrhea have simultaneously trichomoniasis. (85-88)

1.5.2 Path of infection

Humans are the only host for T. vaginalis. The transmission of T. vaginalis is almost always through sexual contact. Since the pathogen can survive in a warm humid environment for 24 hours, a transmission and contamination by infected objects (bath water, bath sponges, toilet seats etc.) is rare but possible. The incubation time is usually between four days to three weeks. (87,89)

1.5.3 Clinical Pathology

The infection is usually oligosymptomatic, chronic, and often goes unnoticed by the wearer. Women are affected more often than men and spontaneous regression of symptoms often occur. In women T. vaginalis prefers the epithelium of the vagina, urethra, Skene's glands, and less frequently the cervix. Rarely, bladder and ureter are affected. Men usually suffer from an infection of the urethra, rarely of the prostate, epididymis or prepuce. (86,87,90)

1.5.3.1 Genital infection in women with Trichomonas vaginalis

Only about 20 % of patients show an acute vulvovaginitis with thin, frothy, muco-purulent, yellowish discharge, painful itching and dysuria. Under examination, the vaginal wall and cervix are red with capillary proliferation and punctiform hemorrhages, giving the vagina a characteristic granular appearance. More often, the infection is chronic with uncharacteristic symptoms such as intermittend itching, erythema of the vulva, slightly elevated discharge, dyspareunia and increased urinary frequency as a result of less

fulminant inflammatory changes in the mucous membranes of the vagina, cervix and urethra. Therefore it can be difficult to clinically differentiate an infection with T. vaginalis from other STDs. Complications of infection with T.vaginalis in women can reach from PID, endometritis to pyolsalpinx. In pregnant women an infection with T. vaginalis can lead to premature rupture of membranes and preterm delivery. (85-87,90)

1.5.3.2 Genital infection in men with Trichomonas vaginalis

T. vaginalis infections in men are usually asymptomatic and therefore trichomoniasis in men is poorly studied. Urethritis is the most common clinical manifestation. The infection with T. vaginalis rarely occurs in a purulent acuteness, instead it often manifests as a subacute-chronic disease. The symptoms are corresponding to other NGNCUs, in which T. vaginalis as well as M. genitalium, U. urealyticum must be considered. (86,90,91) An ascending infection is very rare, but in some cases prostatitis, epididymitis and infertility after a T. vaginalis infection has been described. (87,90)

1.5.4 Diagnostics

In women the most commonly used diagnostic method is the microscopic examination of swabs from the vaginal vault, cervix and urethra. It is based on the characteristic mobility of T. vaginalis in the preparation. In women, about 75 % of the infections can be diagnosed by microscopic examination, subsequently complemented with cell-culture. (86,87,92) The culture is the gold standard, for which various liquid media and cell lines have been described. It has, depending on the medium, a sensitivity of 80-95%. Since complex culture media are technically demanding, expensive and time-consuming, they are not widely used in the routine. (86,87,92)

The PCR allows a rapid diagnosis and is superior in terms of sensitivity (97 %) and specificity (98 %) to all methods previously mentioned. The high sensitivity of this method allows the detection of specimens from morning urine. Routine serological testing is not recommended, because of the variable and unreliable antibody response. (92,93)

The detection of T. vaginalis in men is generally more difficult than in women, therefore cultures to confirm the suspected diagnosis are essential.

1.5.5 Therapy

The CDC-recommended regimen for a T. vaginalis infection is metronidazole or tinidazole, 2 g as a single dose orally or as an alternative metronidazole 500 mg twice daily for five to seven days. This usually shows a response rate of 90-95%. Sexual partners

should be treated as well as asymptomatic patients who were identified by direct detection or culture. Because of the high rate of reinfection among patients rescreening for T. vaginalis at 3 months following initial infection can be considered. (74) In 2%–5% of cases of vaginal trichomoniasis Low-level metronidazole resistance has been identified. (94)
Women can be treated with 2 g metronidazole in a single dose at any stage of pregnancy as studies have proven. (95) However, according to medis (Medikamenten-Informations-System/ drug database) and the Austian guidlines (OEGSTD), metronidazole is contraindicated in the first Trimester and is only recommended for local therapy.

2 Material and Methods

2.1 Purpose of the study

The following chapters refer to the epidemiological study of the data that were assembled from patients that consulted the STI-clinic of the Department of Dermatology and Venereology, Medical University of Graz in the years from May 2006 to May 2011. Data was obtained from MEDOCS (medical records system) and exclusively refers to cases that were recorded at the STI-clinic. Cases having been reported by outside facilities are not included in this thesis.

The main criteria of the data acquisition included sex, age, date of testing and the infection status regarding Chlamydia trachomatis, Neisseria gonorrhoeae, Trichomonas vaginalis, Ureaplasma urealyticum or Mycoplasma hominis.

Through evaluation of the collected data the state of infection and its distribution according to sex and age within the patient population should be revealed.

2.2 Study population

The overall patient population consisted of 1669 patients. In this retrospective study only patients were included who underwent testing for Chlamydia trachomatis, Neisseria gonorrhoeae, Trichomonas vaginalis, Ureaplasma urealyticum or Mycoplasma hominis from May 2006 until May 2011.

To ensure statistical comparability of the collected data two different observation periods and consecutively two test populations were established. All data collected within the period of time from 05/16/2006 to 05/26/2011 were used for the evaluation of the parameters age and sex and their distribution within the patient population.

For the statistical analysis of the pathogen specific infections only the years from 2007 to 2010 (full 12 months) were taken into account to ensure comparability over the years.

2.3 Data acquisition

The implementation of the study in this thesis was approved by the ethics committee of the Medical University of Graz (EK Number: 26-136 ex 13/14). With the help of the IT department of the KAGES (Krankenanstaltengesellschaft) a list of relevant cases among patients tested for the pathogens of interest at the STI-clinic of the University of Graz

during the period of time - May 2006 to May 2011 - was drawn up. The necessary information of each patient was obtained from the electronic medical records system (MEDOCS).

The following parameters were collected for this work. (Figure 4, 5)

Figure 4 – Patient related determinants for data acquisition

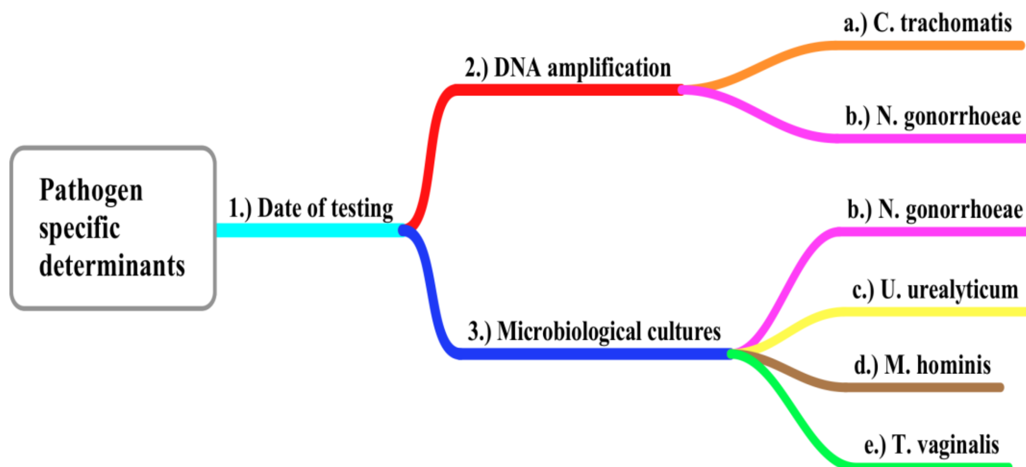


Figure 5 – Pathogen specific determinants for data acquisition

2.4 Data analysis

After having collected the necessary data of MEDOCS, they were integrated and summarized into an Excel file being used as origin for all statistical data analysis with SPSS.

On the other hand these SPSS results were displayed graphically with Excel diagrams.

3 Results

The following chapters refer to the epidemiological study of the collected data for the years 2006 to 2011. This data was obtained from MEDOCS and only contains the cases having been recorded at the STI-clinic of the Department of Dermatology and Venereology Graz.

To ensure statistical comparability of the collected data two different observation periods were established.
All data collected within the period of time from 05/16/2006 to 05/26/2011 were used for the investigation on gender and age related results and their distribution within the patient population.
For the statistical analysis of the pathogen specific results only the years from 2007 to 2010 (full 12 months) were taken into account.

3.1 Gender – related results

3.1.1 All patients tested

1669 patients where tested for C. trachomatis, N. gonorrhoeae, T.vaginalis, U. urealyticum and M. hominis within the period of time from 05/16/2006 to 05/26/2011 by the team of the STI-clinic at the University Clinic for Dermatology and Venereology Graz. Figure 6 exhibits the amount of conducted tests per year, taking into consideration the fact that the statistical evaluation of patients tested in 2006 started in the month of May and ended in the same month in 2011. Therefore only the years from 2007 to 2010 represent the same time interval (12 months) and can be compared statistically.

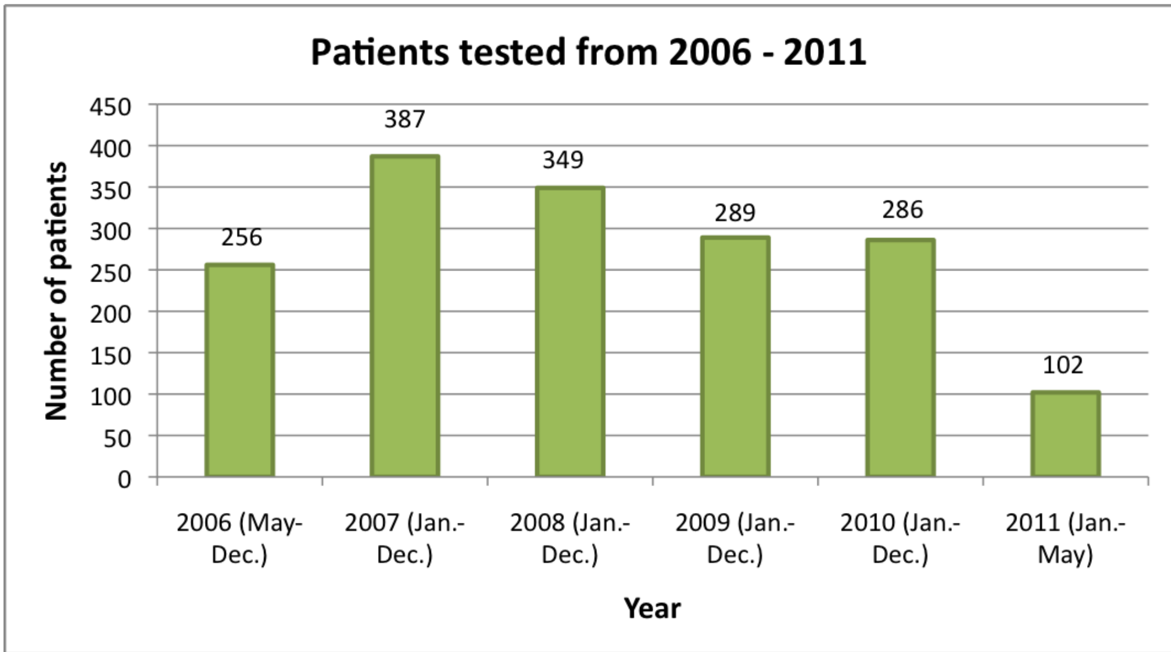


Figure 6 – Patients tested from May 2006 - May2011

Out of all 1669 patients tested 69 % (n = 1159) were of male gender surpassing female patients with 31% (n = 510) by over two-thirds. (Figure 7/8)

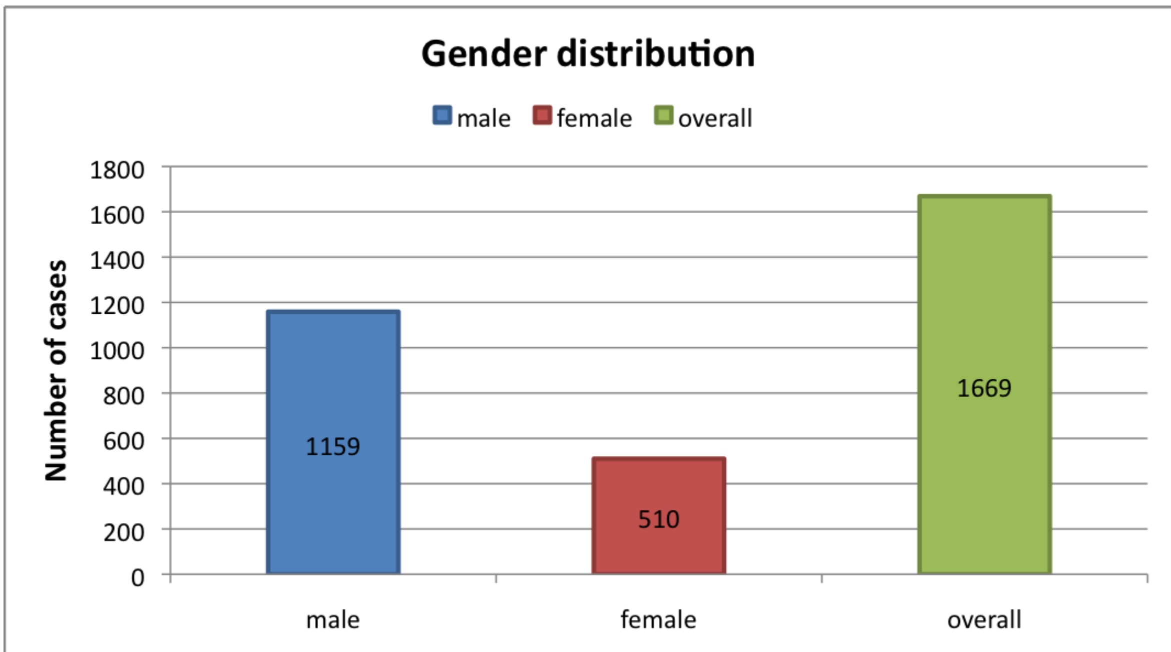


Figure 7 – Gender distribution of all patients tested

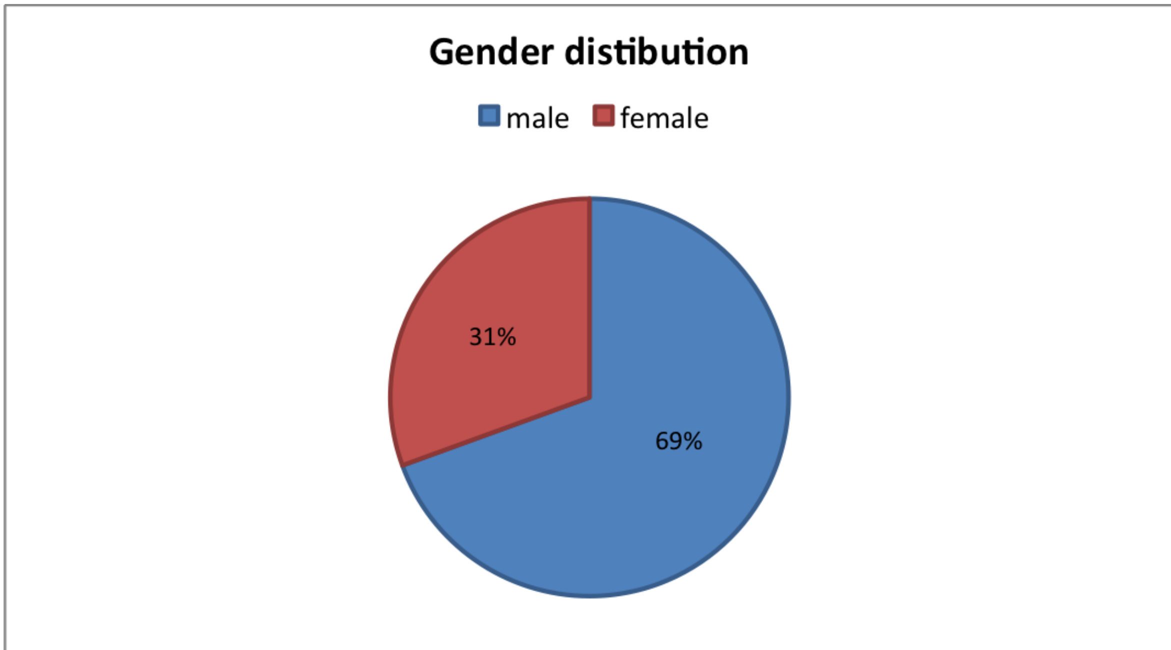


Figure 8 – Gender based percentage of all patients tested

3.1.2 Positive tested patients

Among all of the 1669 patients that underwent examination at the STI-clinic, 39.4% (n= 657) were tested positive for at least one of the pathogens of interest. Looking at the years with full 12 months of testing (2007-2010), most patients tested positive could be found in 2007 (n = 154; 23.4%). (Figure 9/10)

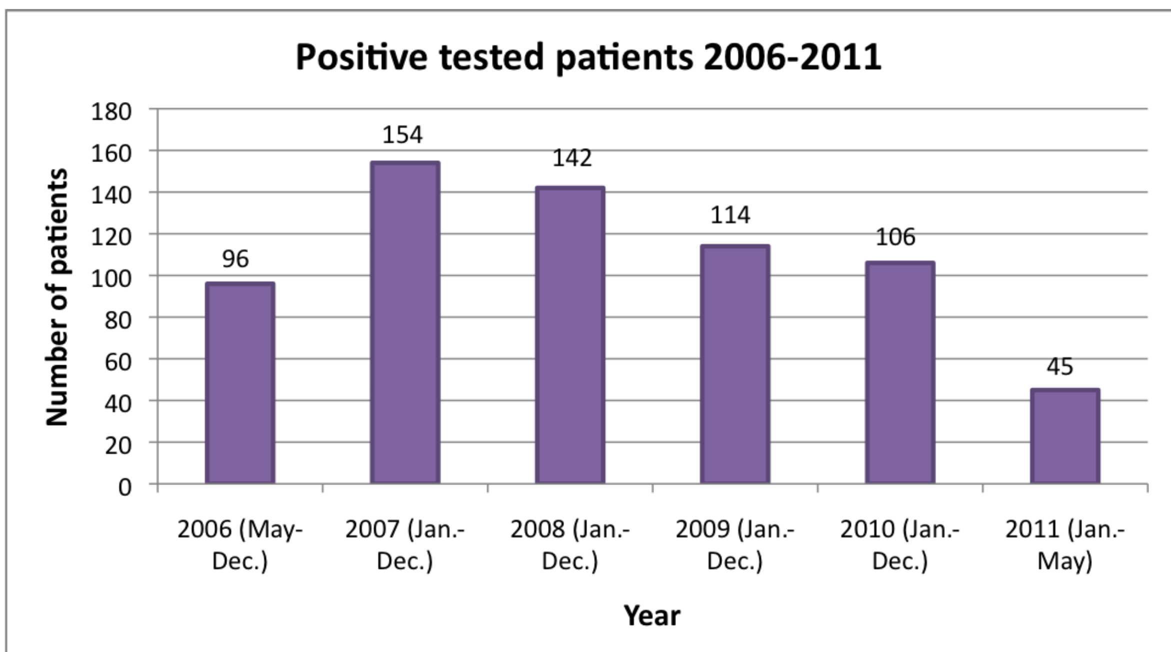


Figure 9 – Positive tested patients May 2006 – May 2011

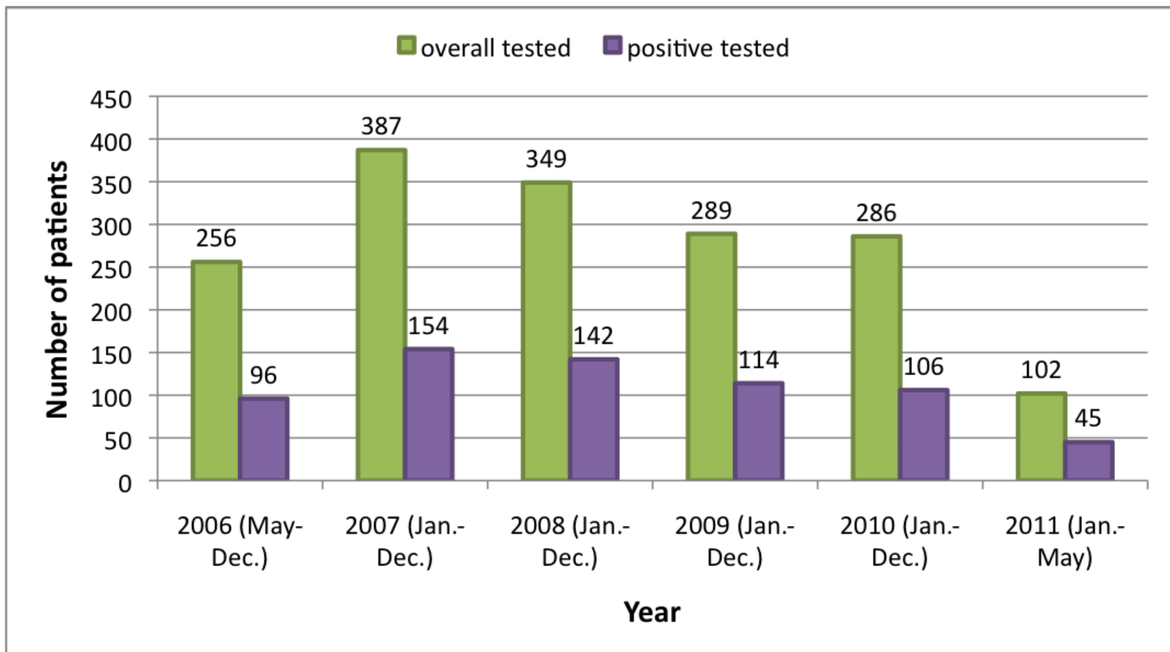


Figure 10 – Overall tested patients vs. positive tested patients

Out of the 657 positive tested patients, 66 % (n = 431) were of male, 34% (n = 226) of female gender. (Figure 11/12)

The ratio of affected men (n=431, 66%) to affected women (n= 226, 34%) within the spectrum of positive tested patients (Figure 12) differed by 6%, compared to the gender percentage distribution of all patients tested. (Figure 8)

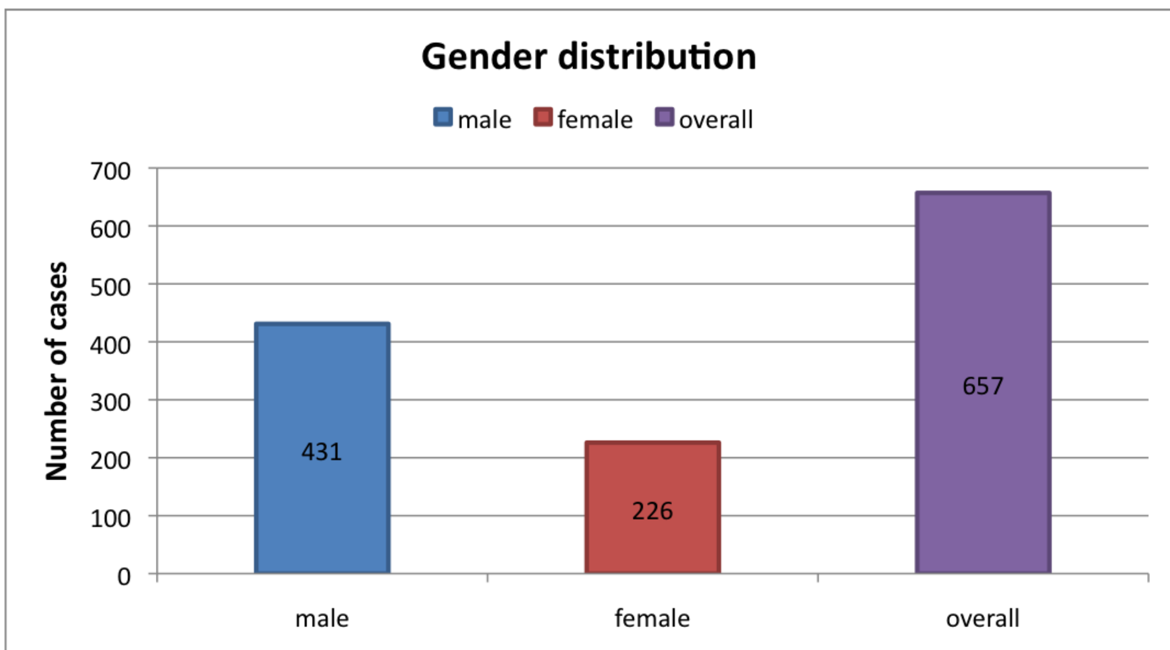


Figure 11 – Gender distribution of all positive tested patients

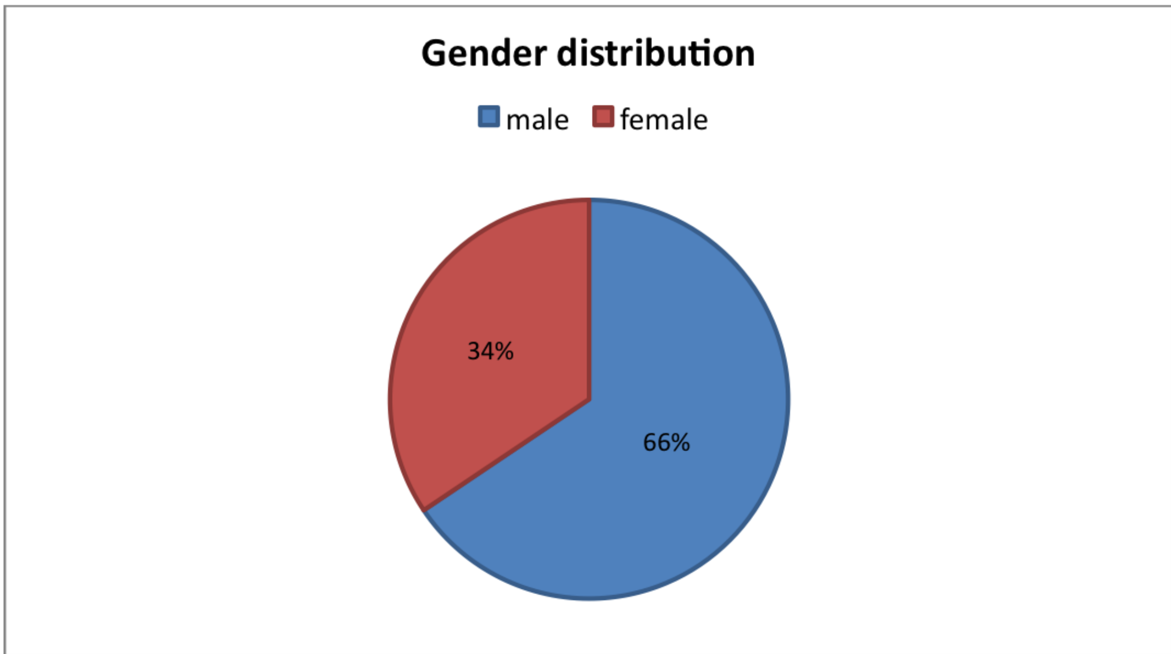


Figure 12 – Gender based percentage of all positive tested patients

As shown by figure 13 the overall percentage of positive tests per year were declining from a first peak in 2008 (40.6%) and rose to a new high in 2011 (44.1%).

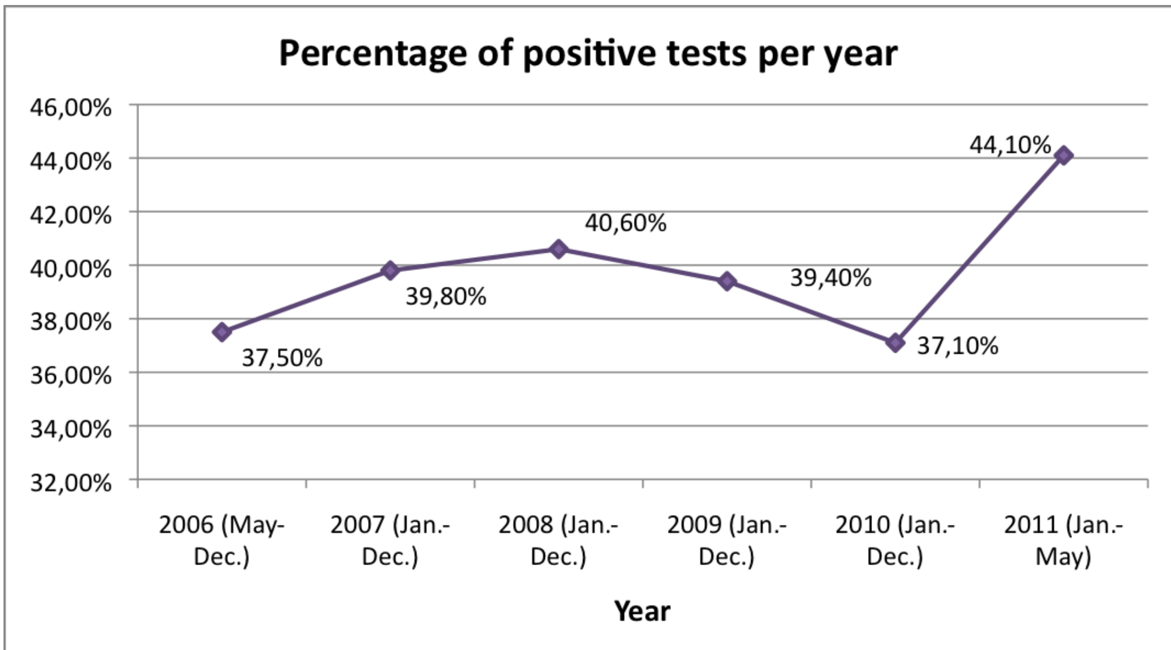


Figure 13 – Overall percentage of positive tests per year

Regarding men, 2007 (39.2%) was the year with the highest percentage of patients tested positive. Four years later in 2011, 59.38% marked the highest percentage of affected female patients. (Figure 14)

Considering the overall number of tested patients per gender, versus the number of persons tested positive, female patients were proven positive in 46.1%, while male patients were identified positive in only 37.1% of all cases, creating a difference of 9%.

In 2006, the difference in percentage between proven positive men in comparison to positive women was only 1.65%. 2007 presented an almost similar number with a difference of 1.68%.

Starting in 2008 (7.71%) and continuing in 2009 (7.68%) the statistical gap became wider with relatively more positive tested females per year. Another leap occurred in 2010 (13.02%) and by 2011 the disparity between the percentage of positive tested men to positive tested women reached its maximum with 22.24%. (Figure 14)

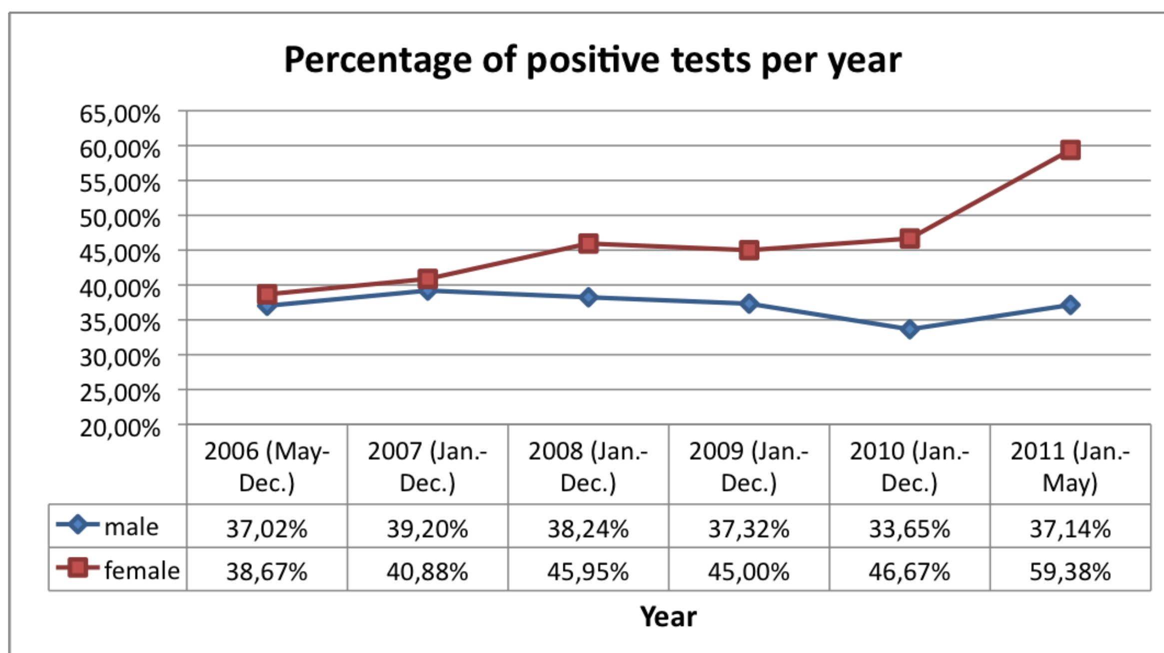


Figure 14 – Percentage of positive tests per year and gender

3.2 Age – related results

3.2.1 All patients tested

At the time of examination the average age in male patients was at 35.7 ± 12.4 years and in female patients at 37 ± 13.7 years. Due to the higher number of male patients tested - who on average were 1.3 years younger than the female patients, the overall mean age at the time of testing was at 36.1 ± 12.8 years.

Parameters regarding distributions in the patients age at the time of STI - testing are shown in figure 15 to 19.

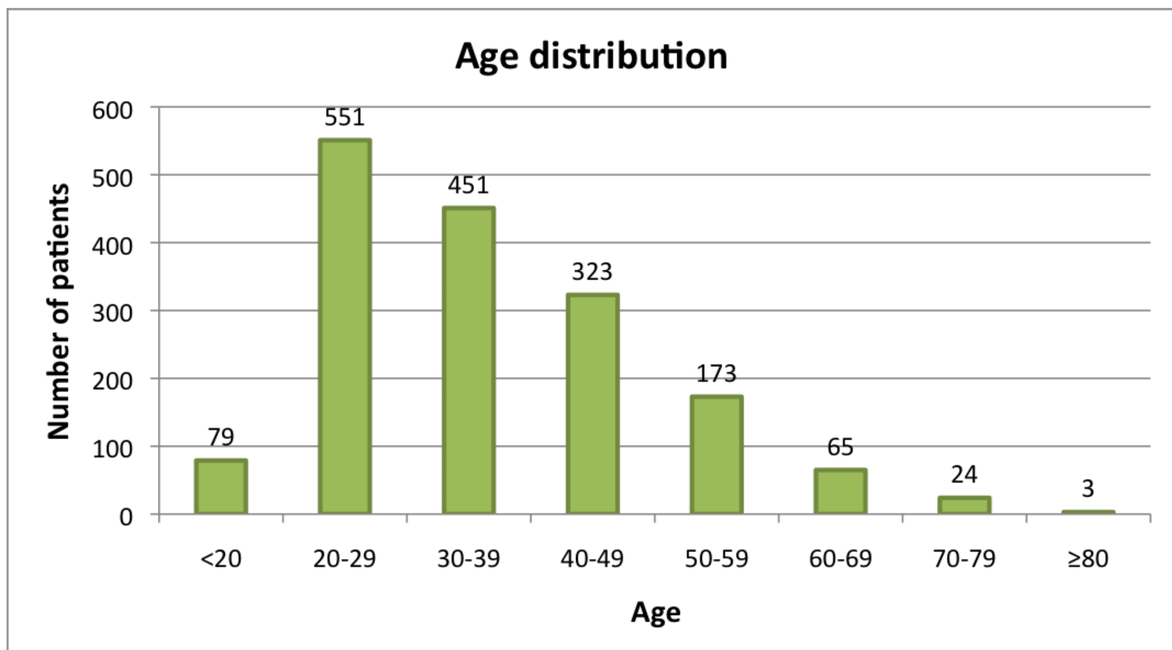


Figure 15 – Age distribution of all patients tested

Patients between the age of 20 and 29 years represented the most common age group in both sexes. Among men 33.8% (n = 392) and 31.1% of women (n = 159) were between 20 and 29 years old.

In both sexes the youngest patient examined was 15 years. The oldest male patient was 79 years old, surpassed by the oldest female patient with an age of 81 years.

Furthermore it can be observed, starting in the patients early 30's the number of consultations at the STD clinic was decreasing that in both sexes.

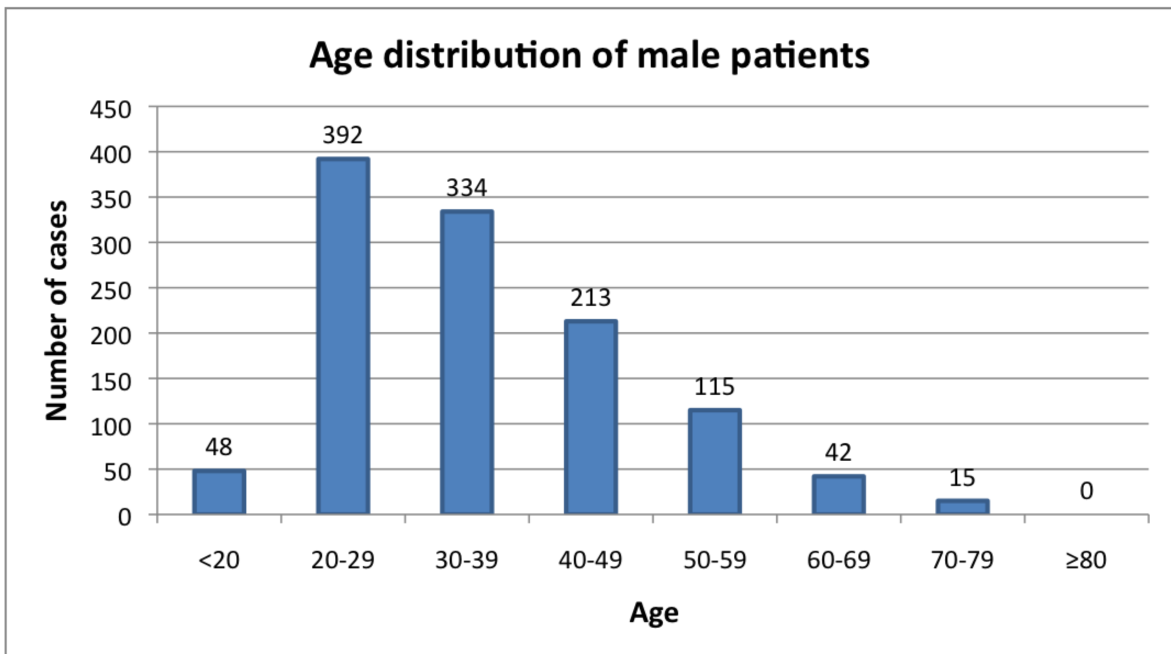


Figure 16 – Age distribution of male patients tested

Under 40-year-old males (n = 774, 66.8%) were tested more frequently for STI-pathogens than patients past the age of 40 years (n = 385, 33.2%).

Bucking the trend, men between the age of 50 and 59 years were more likely to visit the STD clinic (n = 115) than all males under the age of 20 years (n = 48, 4.1%). Under 20 years old patients represented almost the same percentage as men between the ages of 60 to 69 years. (n = 42, 3.6%).

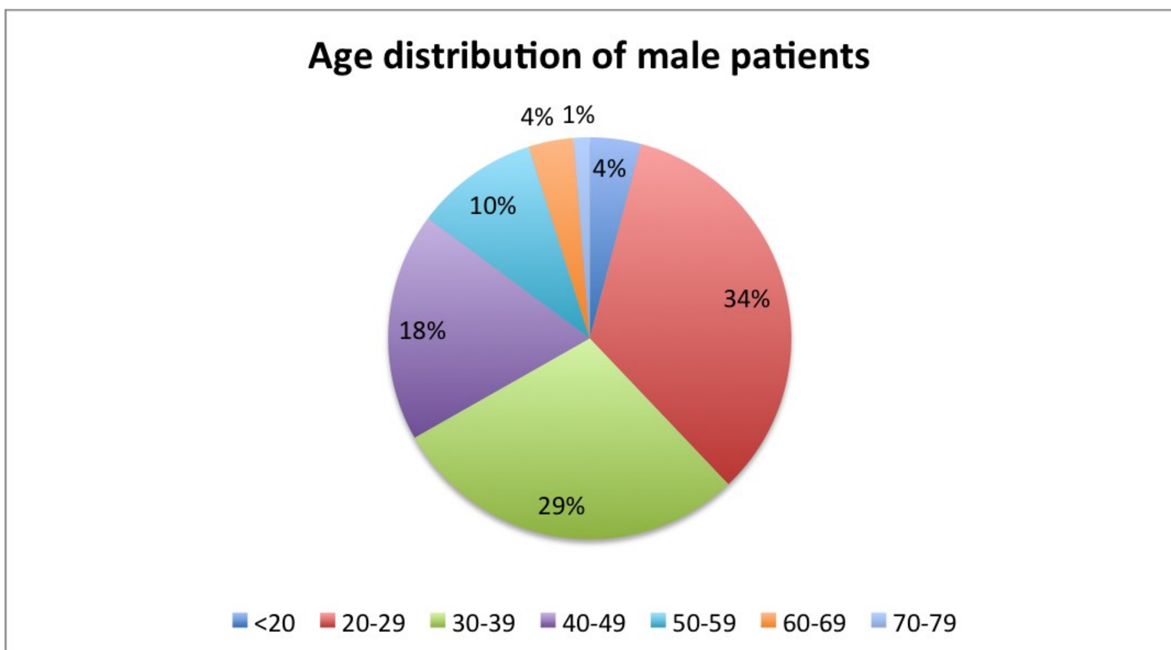


Figure 17 – Age distribution of male patients tested

In women, the empirical study showed a flatter distribution curve than in men.

Still the group of under 40 years old patients (n=307, 60.2%) was represented more often than all the patients passed 40 years (n=203, 39.8%), but the margin between the ages became shorter than in men.

However, the STD clinic was still visited by more female patients between the ages of 50 – 59 (n = 58, 11.4%) than women under the age of 20 (n = 31, 6.1%).

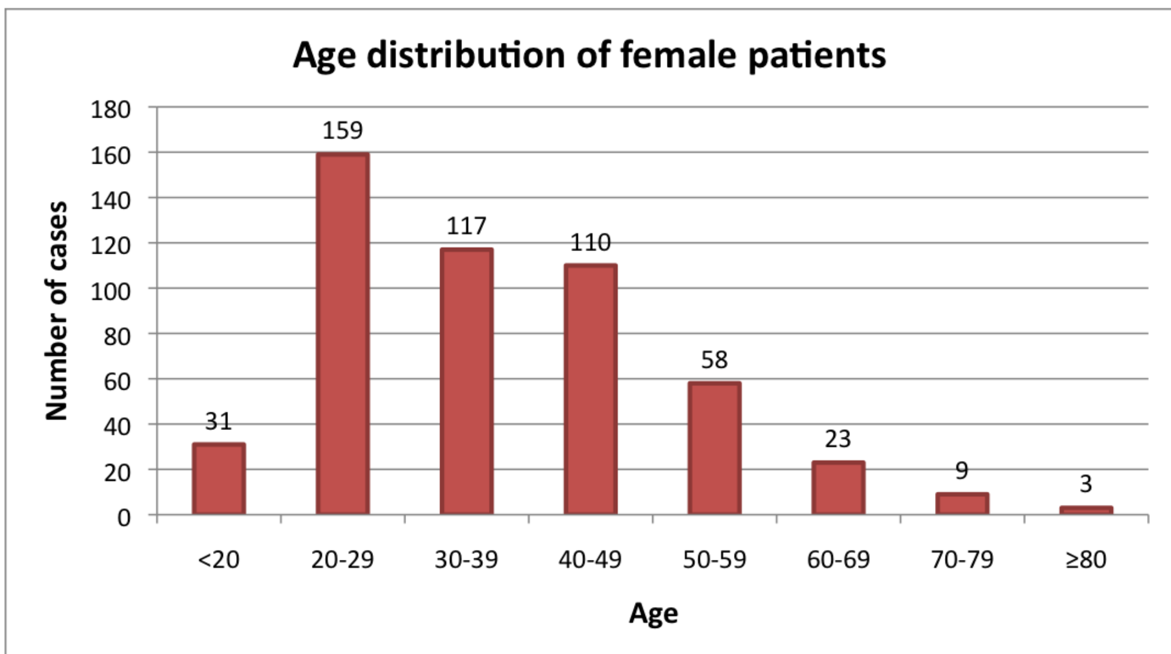


Figure 18 – Age distribution of female patients tested

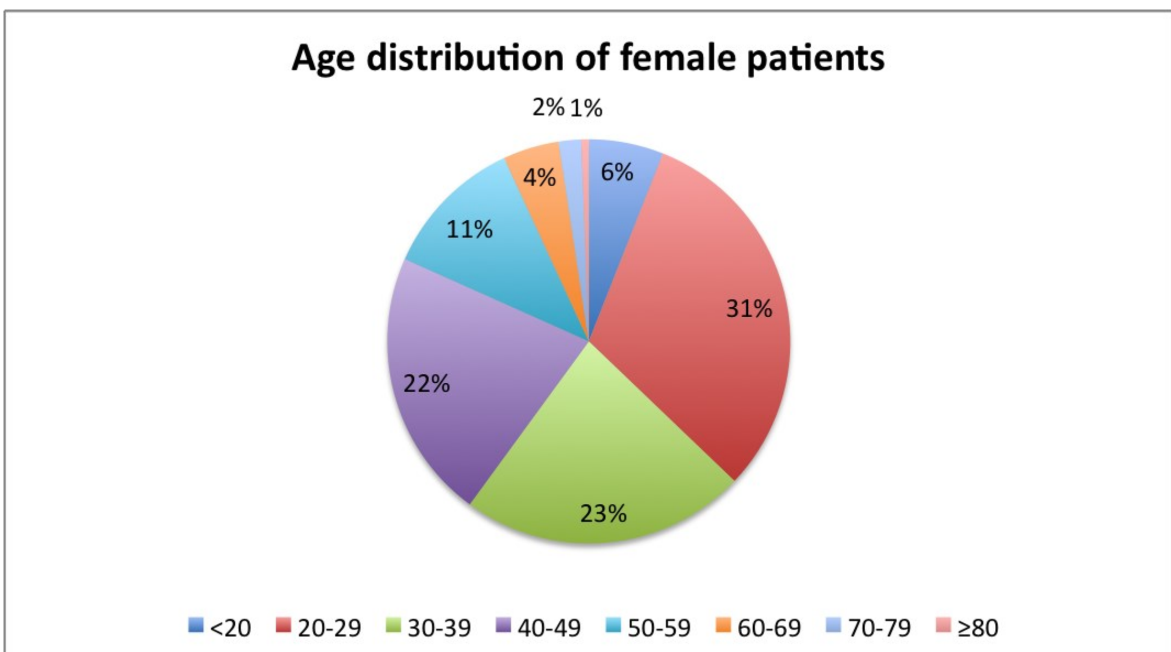


Figure 19 – Age distribution of female patients tested

Comparing the data of both sexes within their age groups, a clear pattern becomes visible. (Figure 20)

Men between the ages of 20 – 39 (n=726) (fig. 16) accounted for 62.6% (fig. 17) of all men tested. Women in the same range of age (n= 276) (fig. 18) only represented 54.1% (fig. 19) of their gender.

Turning to the individual age groups, only in these two age groups (20-29 years and 30-39 years) the percentage of men getting tested was higher than in women. In every other specfic age segment more female patients underwent tests at the STI – clinic than men.

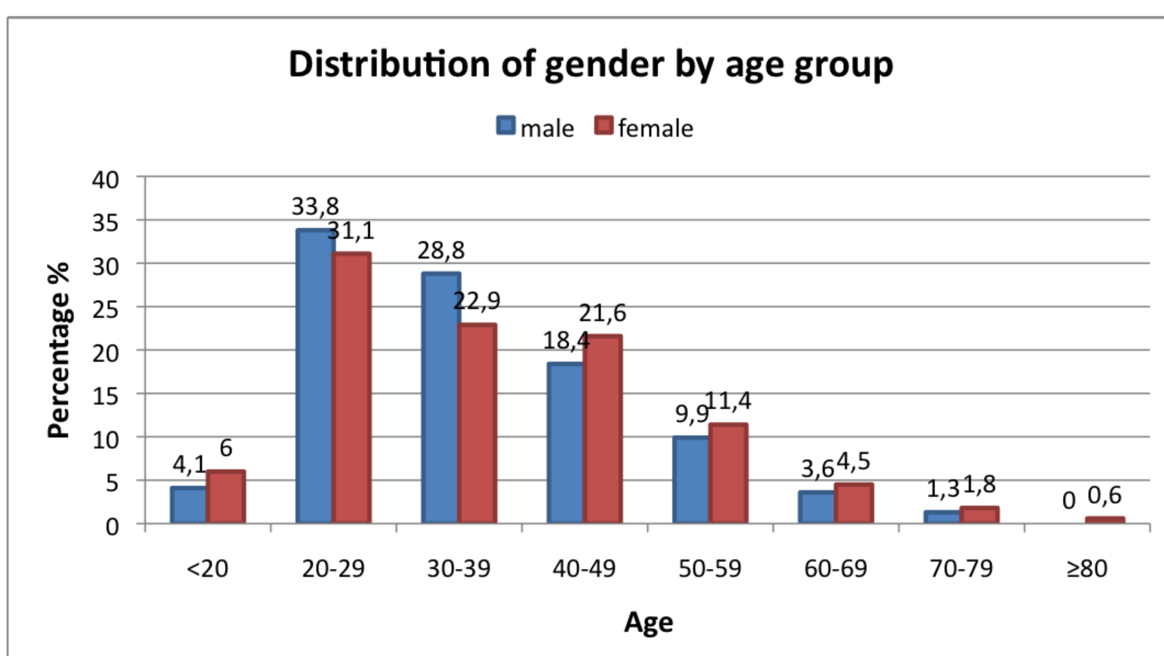


Figure 20 – Distribution of gender by age group of all patients tested

3.2.2 Positive tested patients

The average age in patients tested positive for at least one of the pathogens searched for, differed from the whole tested patient population.

At the time of examination the average age in all male patients tested was at 35.7 years. In positive tested men it marked at 31.1 years, a difference of 4.6 years or 12.8%. Positive tested women (average age 33.3 years) statistically were 3.7 years (10%) younger than the mean age of all females tested (average age 37 years).

The gap between the average ages of men to women increased from 1.3 years (regarding all patients) to 2.2 years (regarding patients tested positive). Influenced by the bigger number of men the overall mean age of positive tested patients was at 31.8 years.

The age distribution of patients tested positive for STI-pathogens is shown in figures 21 to 25.

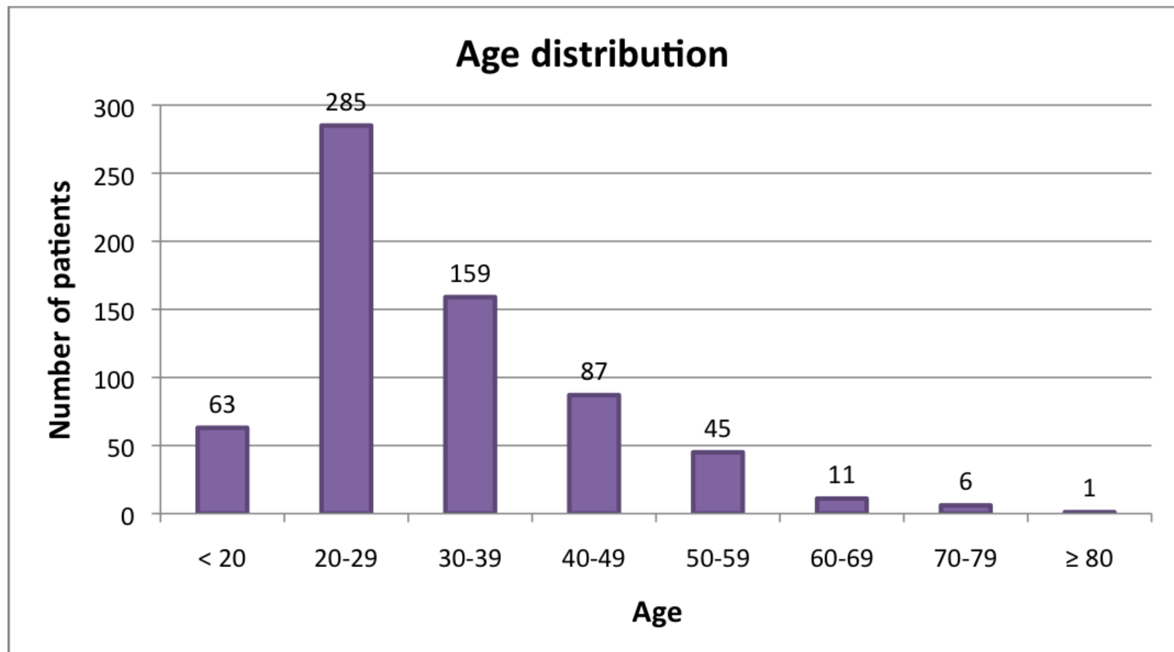


Figure 21 – Age distribution of all positive tested patients

Still Patients between the ages of 20 to 29 years represented the most common age group in both sexes. Among men the percentage has risen significantly about 12.6%, from 33.8% in all patients tested to 46.4% (n= 200) in only positive tested patients. The percentage increase in women between 20 and 29 years old was less significant with 6.5% from 31.1% in all patients tested to 37.6% (n= 85) in only positive tested patients.
The youngest patient tested positive for an STI in both sexes was 16 years, the oldest male patient 78 years and female patient 81 years old.

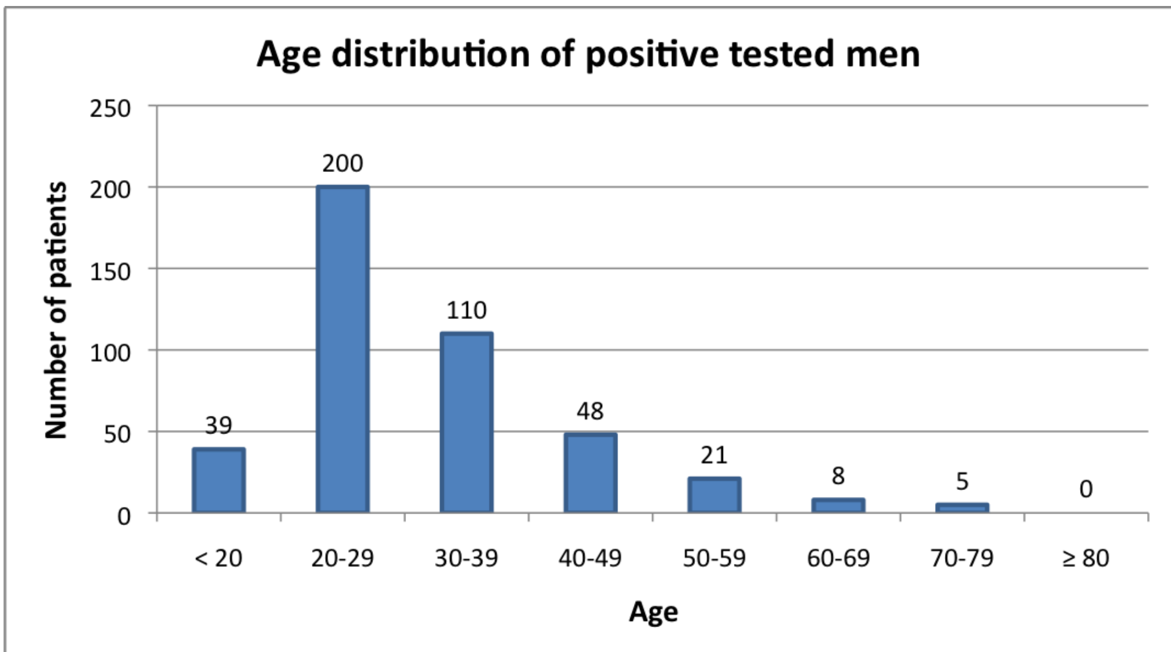


Figure 22 – Age distribution of all positive tested men

The percentage of positive tested men under 40 years rose from 66.8% to 80.9% (n=349). In other words, three out of four tested positive males were under the age of 40 years old. As shown before in figure 16 and figure 20, it was more likely for men between the ages of 50 to 59 years to visit the STD clinic and get tested, than for all males under the age of 20. There was a significant change within in these two age groups of men among the tested positive in comparision to the overall tested. The group of under 20 years old (n= 39.9%) surpassed the group of 50-59 years old men (n=21, 4.9%)

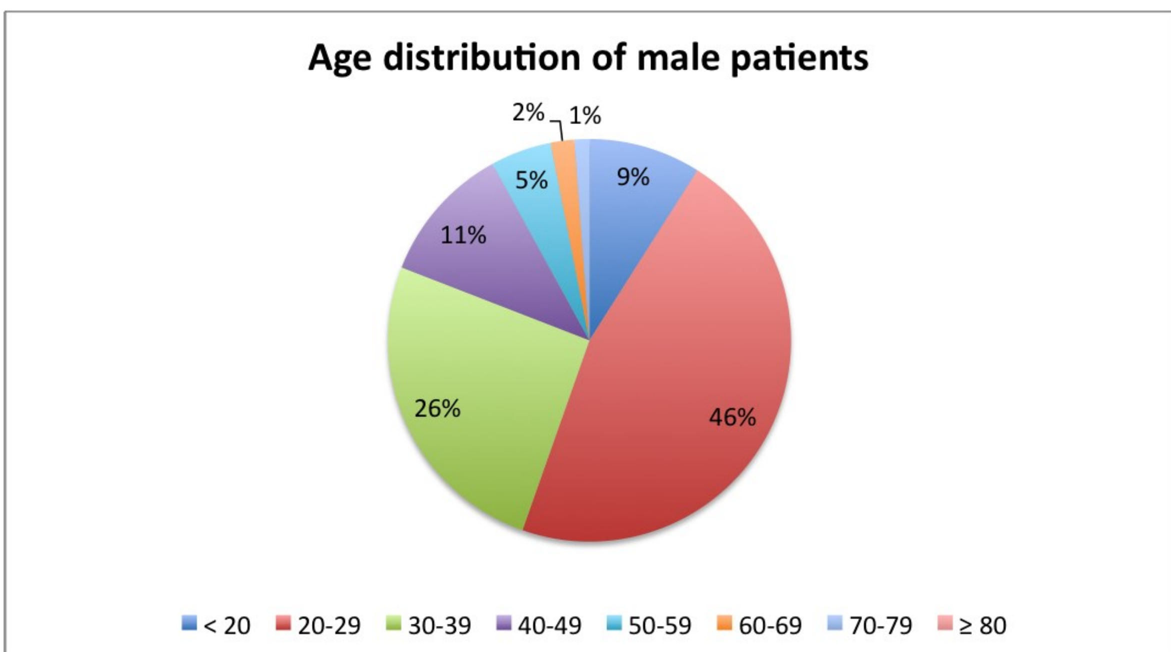


Figure 23 – Percentage distribution of all positive tested men

As shown before in the tested positive male poulation, likewise the women under 40 years were represented more often than all the patients passed 40 years. Still a flatter distribution curve remained within the age groups of the female gender.

The age group of women under 40 years gained 9.7% from 60.2% in all female patients tested, and made up almost 70% of the whole tested positive female patient population.

Contrary to men, female patients between the age of 50 – 59 years (n =24; 11%) and women under the age of 20 years (n = 24; 11%) accounted for the same amount of tested positive patients.

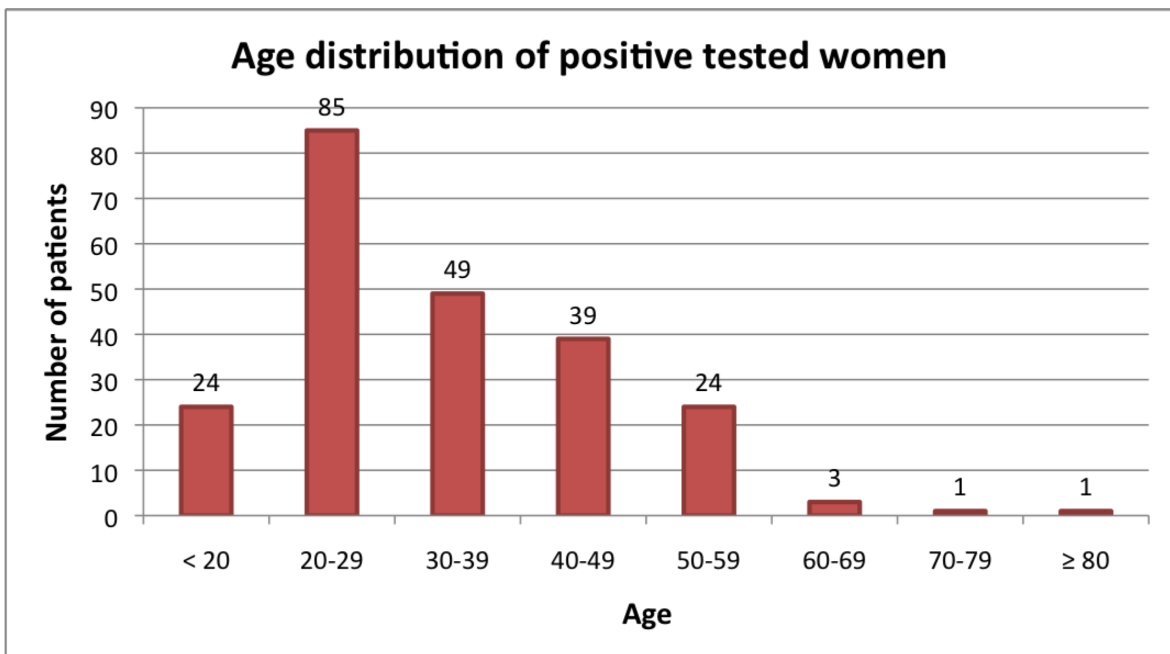


Figure 24 – Age distribution of all positive tested women

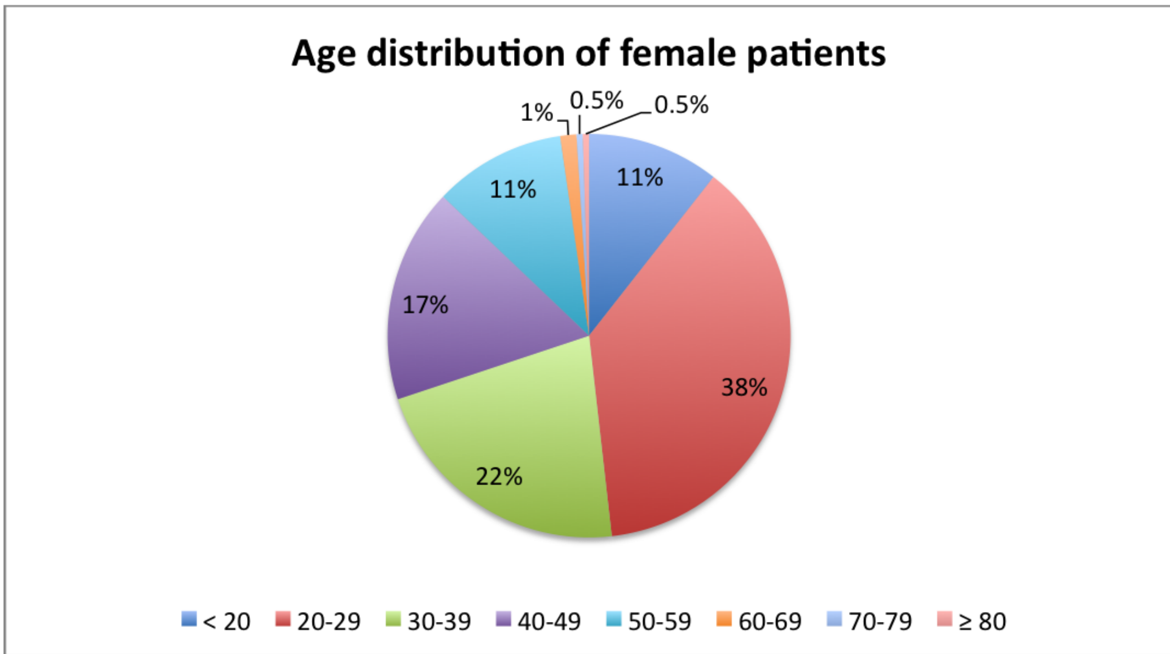


Figure 25 – Percentage distribution of all positive tested women

The former described pattern for the whole tested patient population (figure 20), that only men between the ages of 20 – 39 years have a higher percentage (are more likely to get tested) than women within their age groups, can with small exceptions also be transferred to the positive tested patient population.

The ages of 20 – 39 years not only remained the number one age group within positive tested male and female patients, but also gained 9.4% from 62.6% (fig. 20) to 71.9% (n=310) (fig. 26) in men. The percentage of positive tested women in the same range of age (n= 134), also increased, but only by 5.1% from 54.1% (fig. 20) to 59.2% (fig. 26) of their gender.

Exceptions of the former mentioned pattern can be found within the individual age groups. In the overall patient population it was only in the two age groups from 20-29 years and 30-39 years that the percentage of men tested was higher than in women and in every other specific age segment women were more likely to get tested.

This changed in the distribution of all positive tested patients by age group. (fig. 26)

In the age groups of 60-69 years and 70-79 years the percentage of positive tested men overlapped the percentage of positive tested women within the genders (fig. 22/24), both sexes loosing percentages to younger age groups.

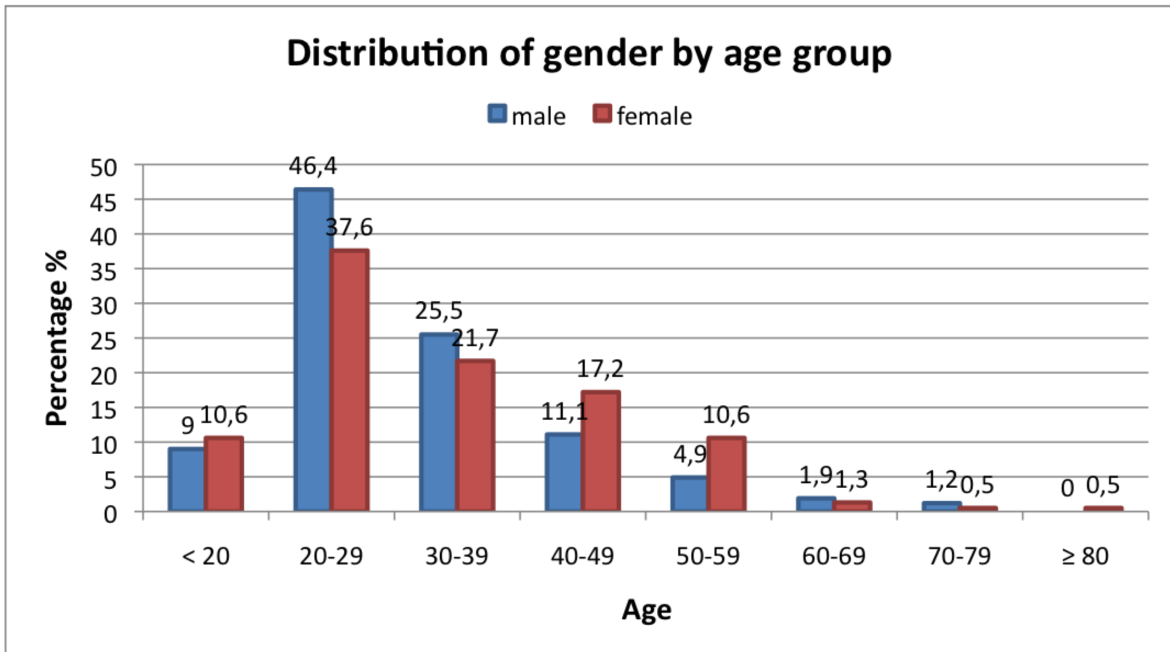


Figure 26 – Distribution of gender by age group of all positive tested patients

Table 3 illustartes an overview of the collected data regarding the age distribution of the study population from 2006 until 2011.

2006-2011	Study population n=1669			Positive cases n=657		
Age group	**Men**	**Women**	**Total**	**Men**	**Women**	**Total**
<20	48 (4.1%)	31 (6%)	79 (4.7%)	39 (9.0%)	24 (10.6%)	63 (9.6%)
20 - 29	392 (33.8%)	159 (31.3%)	551 (33.0%)	200 (46.4%)	85 (37.6%)	285 (43.4%)
30 - 39	334 (28.8%)	117 (22.9%)	451 (27.0%)	110 (25.5%)	49 (21.7%)	159 (24.2%)
40 - 49	213 (18.4%)	110 (21.6%)	323 (19.4%)	48 (11.1%)	39 (17.3%)	87 (13.2%)
≥50	172 (14.8%)	93 (18.2%)	265 (22.7%)	34 (7.8%)	29 (12.8%)	63 (9.6%)

Table 3 – Study population/ Age distribution

3.3 Pathogen specific results

This chapter will discuss the distribution of pathogens of interest as well as their separate numbers within the sexes. (Figure 28, 29 and Table 5)

To ensure statistical comparability only the years from 2007 to 2010 (full 12 months) were taken into account. (Figure 27)

Emanating from the entity in all positive tested patients (n= 516) in these years, table 4 exhibits their dispersal within the genders.

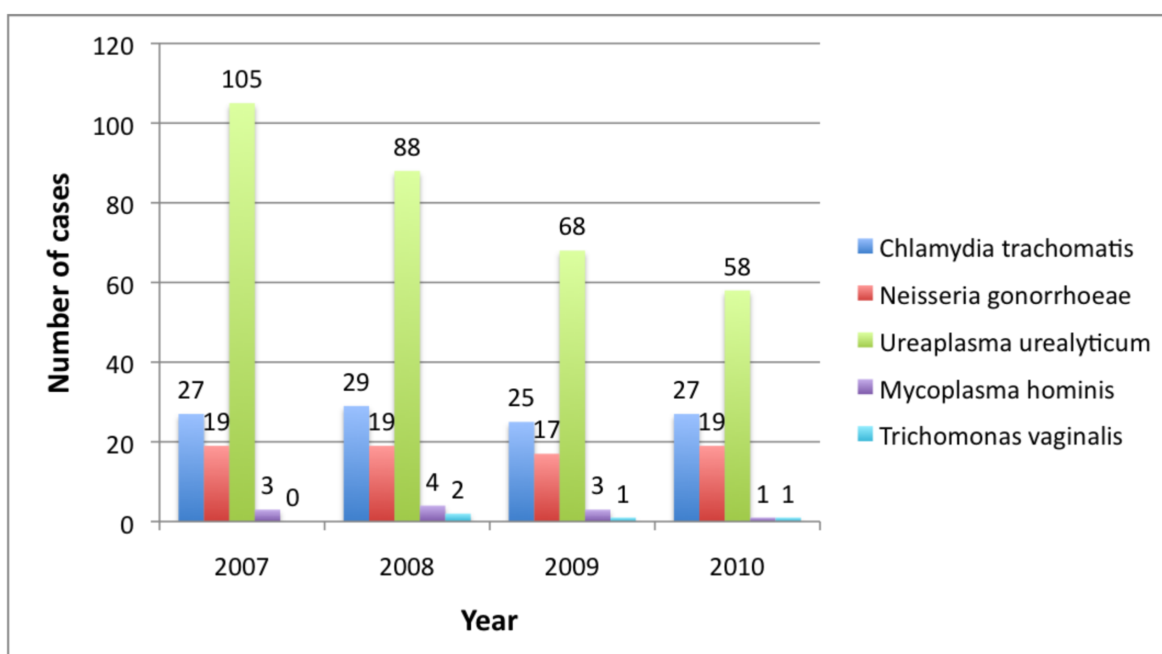


Figure 27 – Distribution of pathogens from January 2007 to December 2010

	male	%	female	%	overall	%
Chlamydia trachomatis	94	27.8	14	7.9	108	20.9
Neisseria gonorrhoeae	66	19.5	8	4.5	74	14.5
Ureaplasma urealyticum	173	51.2	146	82.0	319	61.8
Mycoplasma hominis	4	1.2	7	3.9	11	2.1
Trichomonas vaginalis	1	0.3	3	1.7	4	0.8
Σ	338		178		516	

Table 4 – Distribution of pathogens

Both sexes displayed the same hierarchy within the pathogens. The most common pathogen found in male (n= 173; 51.2%) and female (n= 146; 82.0%) patients was U. urealyticum. Followed by C. trachomatis (m.: n=94; 27.8% / f.: n=14; 7.9%), N. gonorrhoeae (m.: n=66; 19.5% / f.: n=8; 4.5%), M. hominis (m.: n=4; 1.2% / f.: n=7; 3.9%) and T. vaginalis (m.: n=1; 0.3% / f.: n=3; 1.7%).

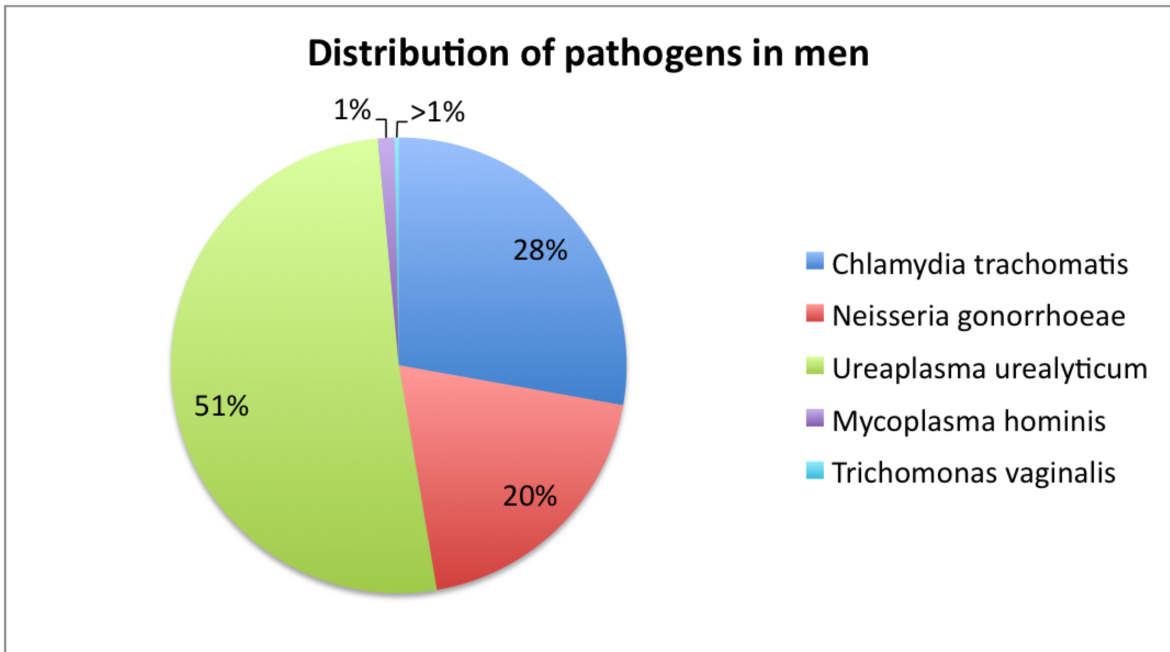


Figure 28 – Distribution of pathogens in men

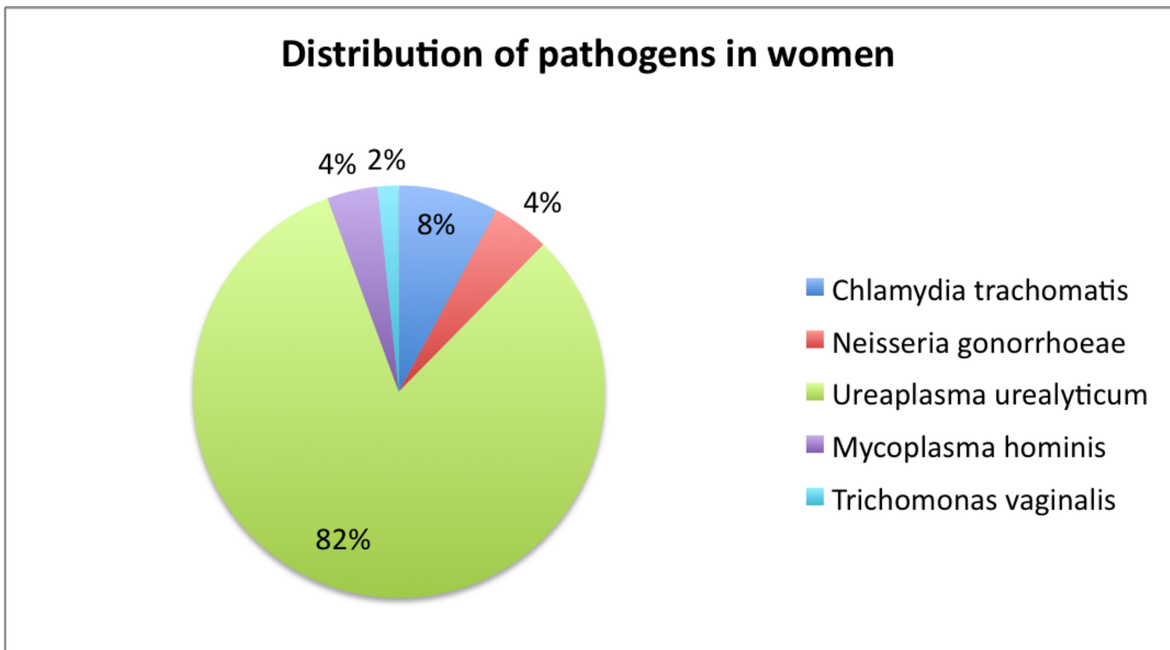


Figure 29 – Distribution of pathogens in women

2007-2010	<20	20 - 29	30 - 39	40 - 49	≥ 50	Total
C. trachomatis cases Graz	14 (13.0%)	51 (47.2%)	29 (26.9%)	8 (7.4%)	6 (5.5%)	108 (20.9%)
N. gonorrhoeae cases Graz	7 (9.5%)	32 (40.5%)	19 (24.1%)	9 (12.2%)	7 (9.5%)	74 (14.4%)
U. urealyticum cases Graz	30 (9.4%)	132 (41.4%)	70 (21.9%)	49 (15.4%)	38 (11.9%)	319 (61.8%)
M. hominis cases Graz	2 (18.2%)	5 (45.4%)	0 (0%)	3 (27.3%)	1 (9.1%)	11 (2.1%)
T. vaginalis cases Graz	0 (0%)	1 (25%)	1 (25%)	1 (25%)	1 (25%)	4 (0.8%)
Total	53 (10.3%)	221 (42.8%)	119 (23.1%)	70 (13.5%)	53 (10.3%)	516 (100%)

Table 5 – Pathogen/ Age distribution

3.3.1 Ureaplasma urealyticum

Being the most common STI–pathogen in the tested patient population, U. urealyticum (n= 319; 61.8%) could be found in both sexes every year in the examined period of time 2007-2010. Male patients accounted for 173 (54.2%) and females for 146 (45.8%) of them.
After peaking in 2007 (n= 105; 68.2%), the number of patients per year kept reducing until it reached its lowest level in 2010 (n= 58; 54.7%). (Figure 30)
2009 showed the year with the biggest difference (20.6%) between positive tested men (n= 41; 60.3%) to women (n= 27; 39.7%). The overall patient number was 68, amounting to 59.6% of all patients in 2009 tested positive for a STI-pathogen of interest.

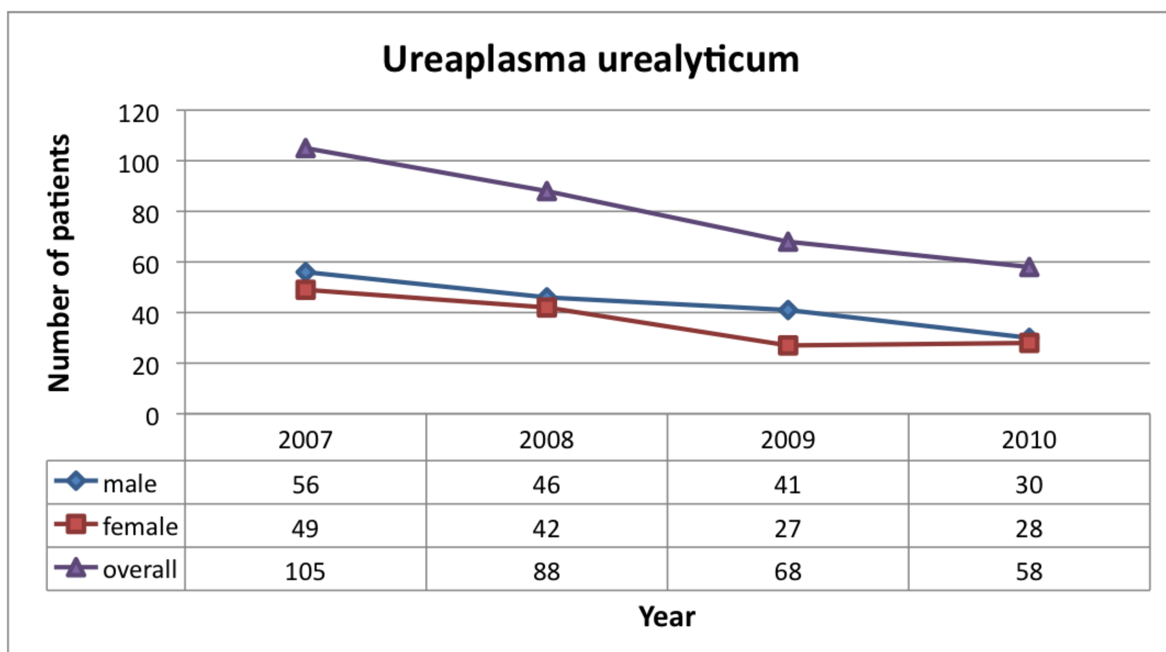


Figure 30 – Gender distribution of Ureaplasma urealyticum

Looking at distribution of age among U. urealyticum positive patients, proportions apparently follow the overall trend. Patients between the ages of 20-29 years accounted for 41.4% (n= 132) out of all the 319 positive tested patients; followed by the age groups of 30-39 years (n= 70; 21.9%), 40-49 years (n= 49; 15.4%), ≥ 50 years (n= 38; 11.9%) and last by the < 20 years (n= 30; 9.4%). Considering the years of testing separately, the positions of youngest to oldest group tested positive get turned around. (fig. 31)
In 2007, the < 20 year olds only represented for 3.8% (n= 4) of all patients tested positive, while 17 patients (16.2%; quadruple the number of < 20 year olds) belonged to the age group of ≥ 50 years old. One year later, in 2008 the STD – clinic detected 13 cases (14.8%) of patients < 20 years old, in comparision to only 10 (11.4%) of ≥ 50 year olds and 9 of 40-49 year old patients. The age group of 20-29 year olds accounted for almost 50% of all positive tested U. urealyticum cases in 2009 and 2010, whilst in 2008 the same age group only marked a third of positive cases.

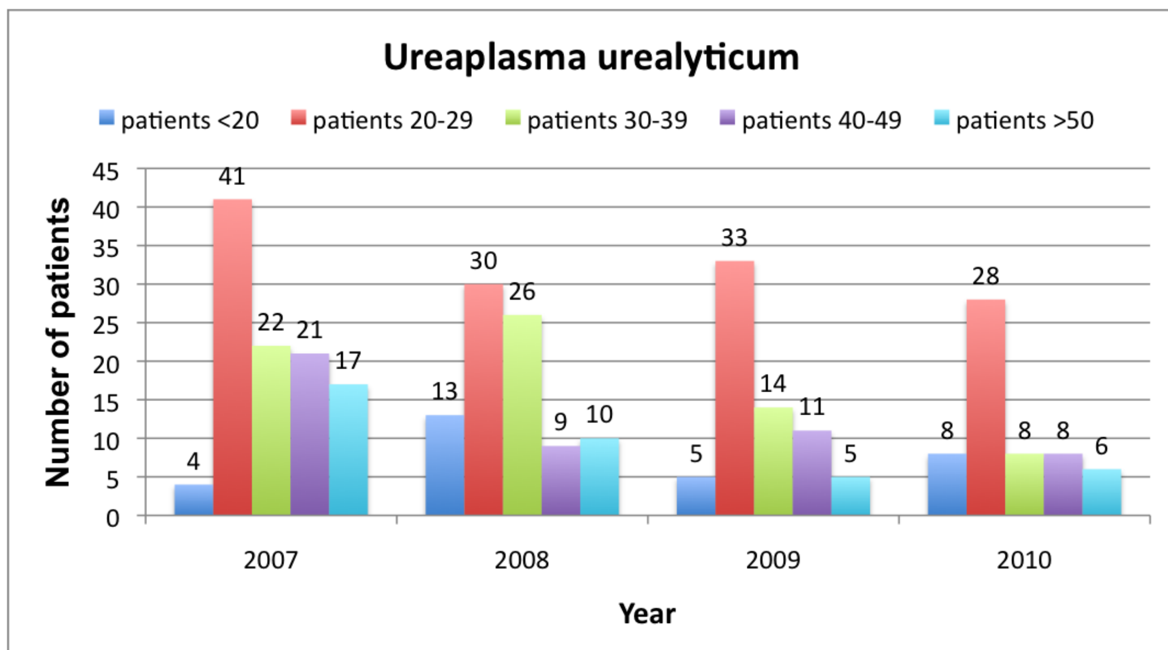


Figure 31 – Age distribution of Ureaplasma urealyticum

3.3.2 Chlamydia trachomatis

Out of a total number of 108 tested positive C. trachomatis cases male patients accounted for 94 (87.0%), females for only 14 (13.0%) of them.
Over the entire test period from 2007-2010 the proportion between the sexes remained the same. 2008 marked the year with the biggest difference (79.4%) between positive tested

men (n= 26; 89.7%) to women (n= 3; 10.3%), with an overall patient number of 29. This marked 20.4% of all patients tested positive for a STI-pathogen of interest in 2008.
It also represented the peak in number of patients per year. (fig. 32)

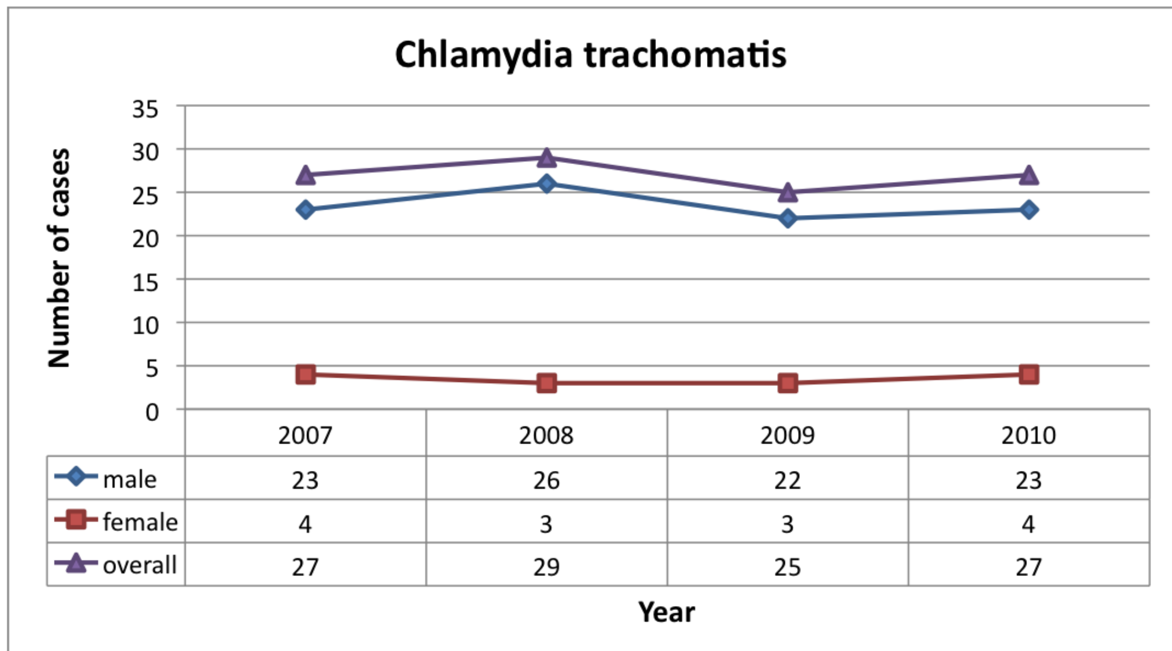


Figure 32 – Gender distribution of Chlamydia trachomatis

In accordance with the general age distribution of all tested positive patients the ages of 20-29 years accounted for more than half (n= 51; 47.2%) out of the 108 positive C. trachomatis cases. Followed again by the age group of 30-39 years (n= 29; 26.9%). Contrary to the age distribution results of U. urealyticum and the overall trend, the < 20 years olds (n= 14; 13.0%) were the third biggest group, succeeded by 40-49 years old (n= 8; 7.4%) and ≥ 50 years (n= 6; 5.5%). (fig. 33)

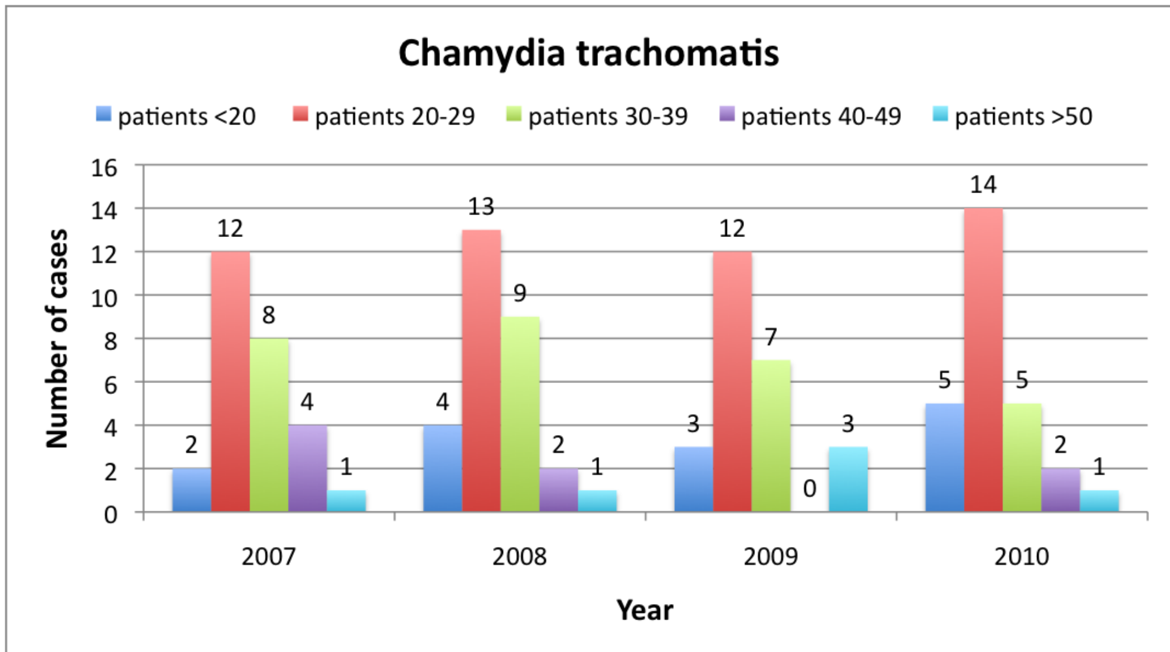


Figure 33 – Age distribution of Chlamydia trachomatis

3.3.3 Neisseria gonorrhoeae

In almost 15% of all positive tested patients N. gonorrhoeae was the pathogen detected and accounted for a combined total of 74 cases from 2007 – 2010. 9 out of 10 patients were of male gender (n= 66; 89.2%). (fig. 34)

Over the entire test period from 2007-2010 the prevalence remained on a constant level. 2007 was the year with the biggest difference (89.4%) between positive tested men (n= 18; 94.7%) to women (n= 1; 5.3%), with an overall patient number of 19, marking for 12.3% of all patients in 2007 tested positive for a STI-pathogen of interest.

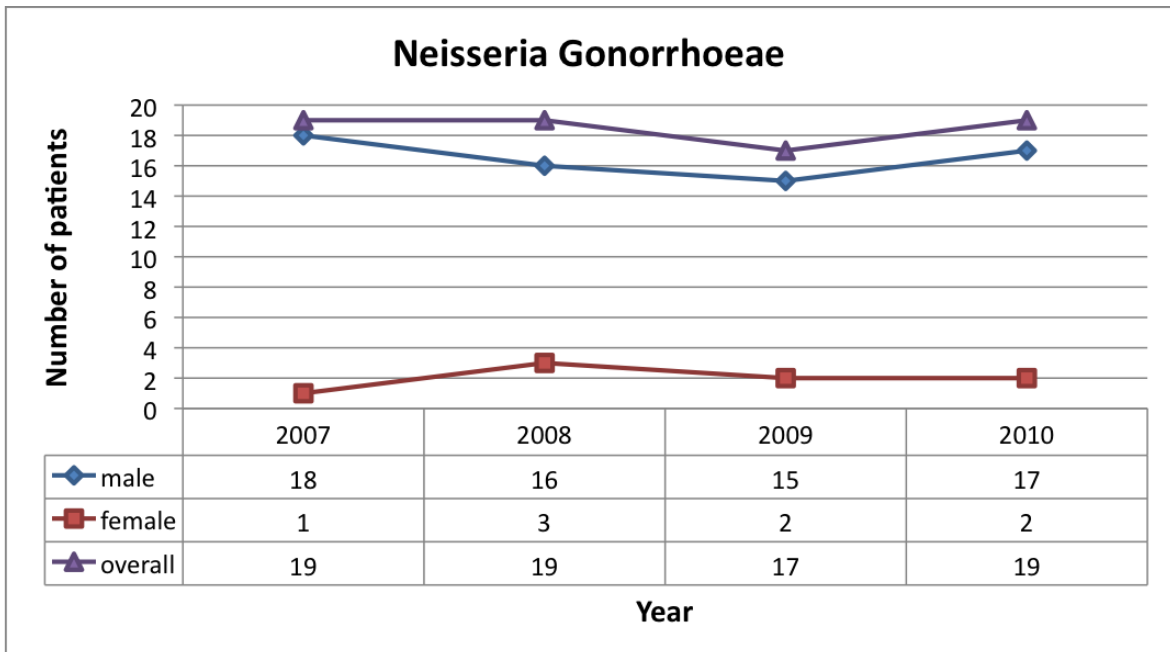


Figure 34 – Gender distribution of Neisseria gonorrhoeae

Within the positive tested N. gonorrhoeae collective age distribution, in its entity, followed the overall trend. Patients between the ages of 20-29 years accounted for 40.5% (n= 32) out of all the 74 positive tested patients; followed by the age groups of 30-39 years (n= 19; 24.1%), 40-49 years (n= 9; 12.2%). Last in line the ages ≥ 50 years (n= 7; 9.5%) and < 20 years (n= 7; 9.5%) showed the same amount of cases. (fig. 35)

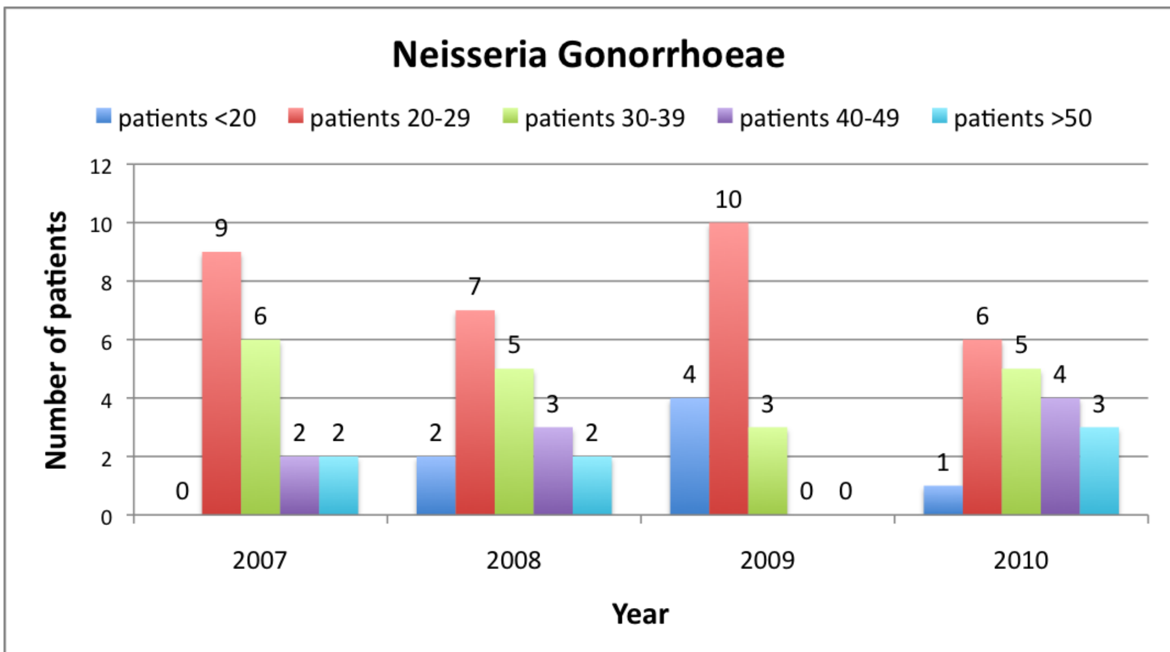


Figure 35 – Age distribution of Neisseria gonorrhoeae

3.3.4 Mycoplasma hominis

M. hominis only accounted for 2.1% (n=11) out of all the positive tested patients. Apparently it was more common in females (n=7; 63.6%) than in males (n=4; 36.4%) referring to the examined population.

Patients of both sexes could be found every year until 2009, when the patient numbers started to decline, reaching its lowest point in 2010 with only one positive male case. (fig. 36)

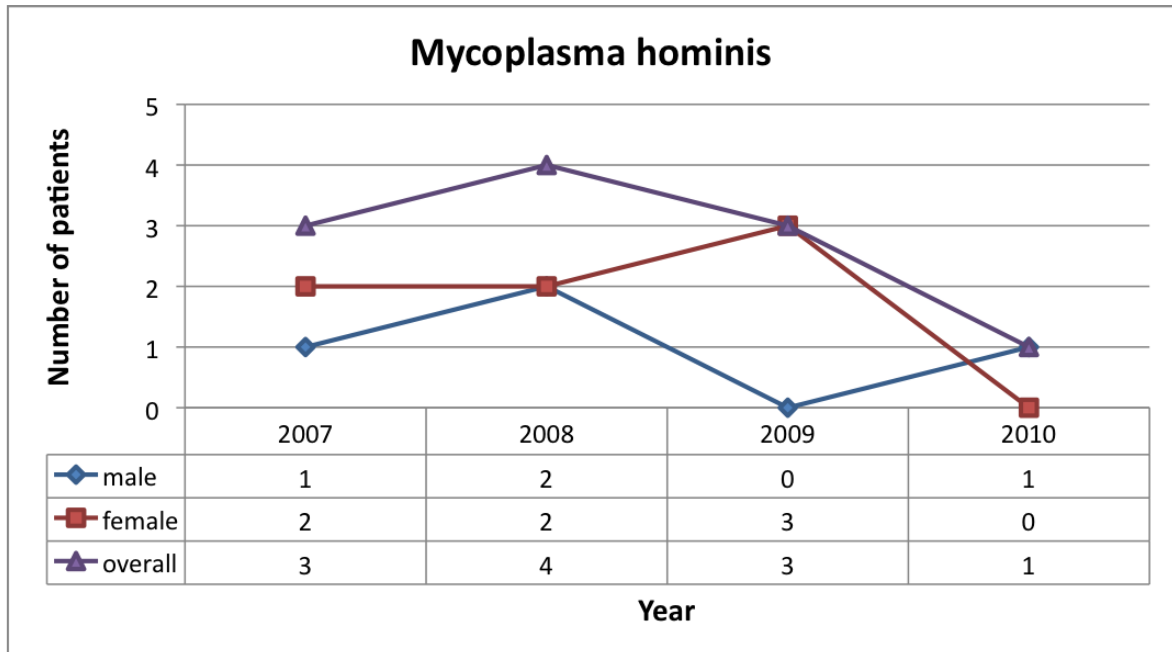


Figure 36 – Gender distribution of Mycoplasma hominis

In contrast to the general age distribution of all positive tested patients there were no cases of patients between the ages of 30-39 years over the whole examined time spectrum.

None the less the group of 20-29 years accounted for 45.4% (n= 5) out of all 11 positive tested patients. Followed by the age groups of 40-49 years (n= 3; 27.3%), < 20 years (n= 2; 18.2%), and last by the ≥ 50 years (n= 1; 9.1%). (fig. 37)

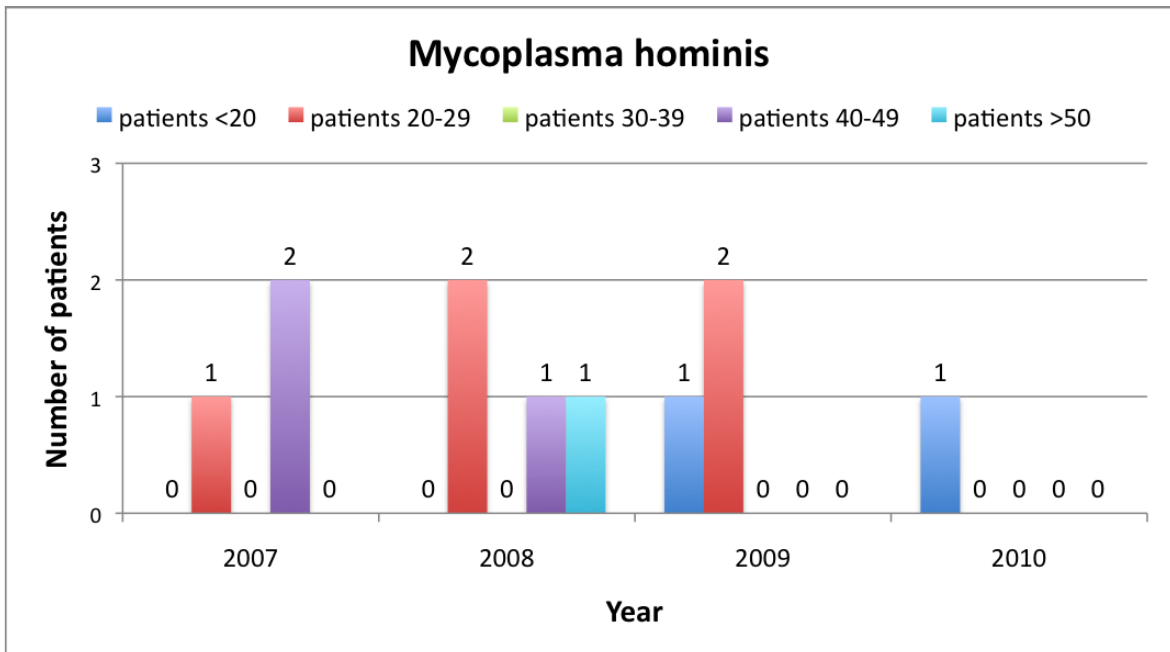
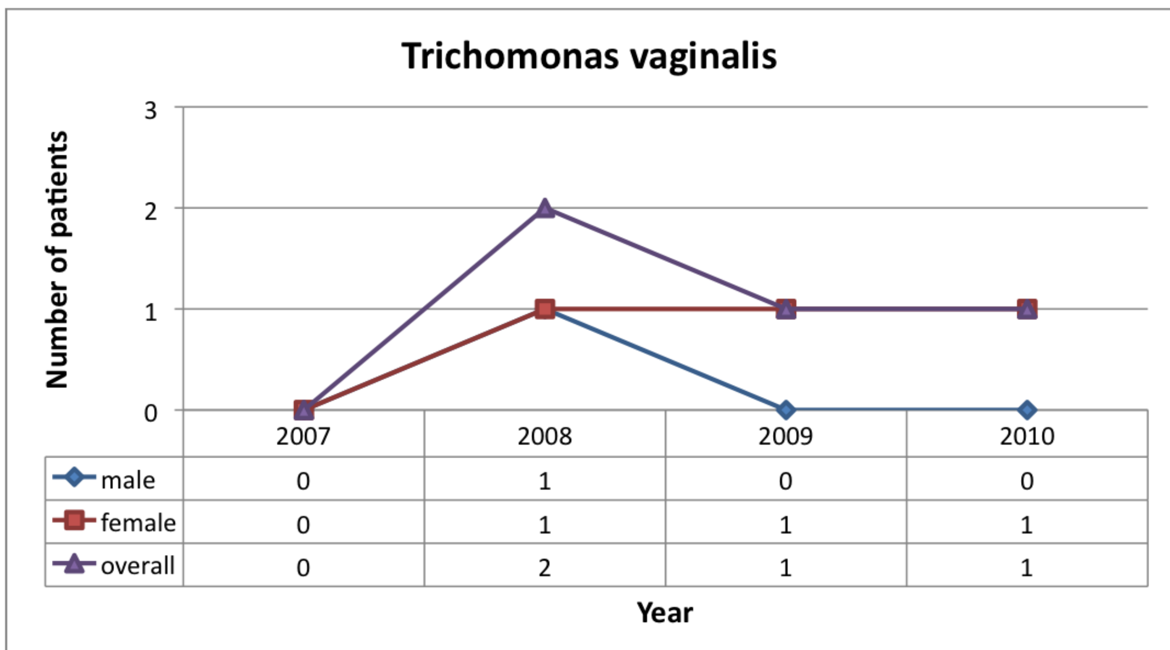


Figure 37 – Age distribution of Mycoplasma hominis

3.3.5 Trichomonas vaginalis

The smallest group within the whole examined germ spectrum at the STD outpatient clinic Graz, with only four positive tested patients, accounted for only 0.8% of all positive cases from 2007 – 2010. Out of these four cases only one male patient was infected, making T. vaginalis the pathogen with the highest percentage (75%) of female patients. (fig 38)



	2007	2008	2009	2010
male	0	1	0	0
female	0	1	1	1
overall	0	2	1	1

Figure 38 – Gender distribution of Trichomonas vaginalis

Owing to the small number of positive cases, the age distribution within the T. vaginalis positive patients was relativly even distributed over all age groups with the exception of no patients < 20 years of age. (fig 39)

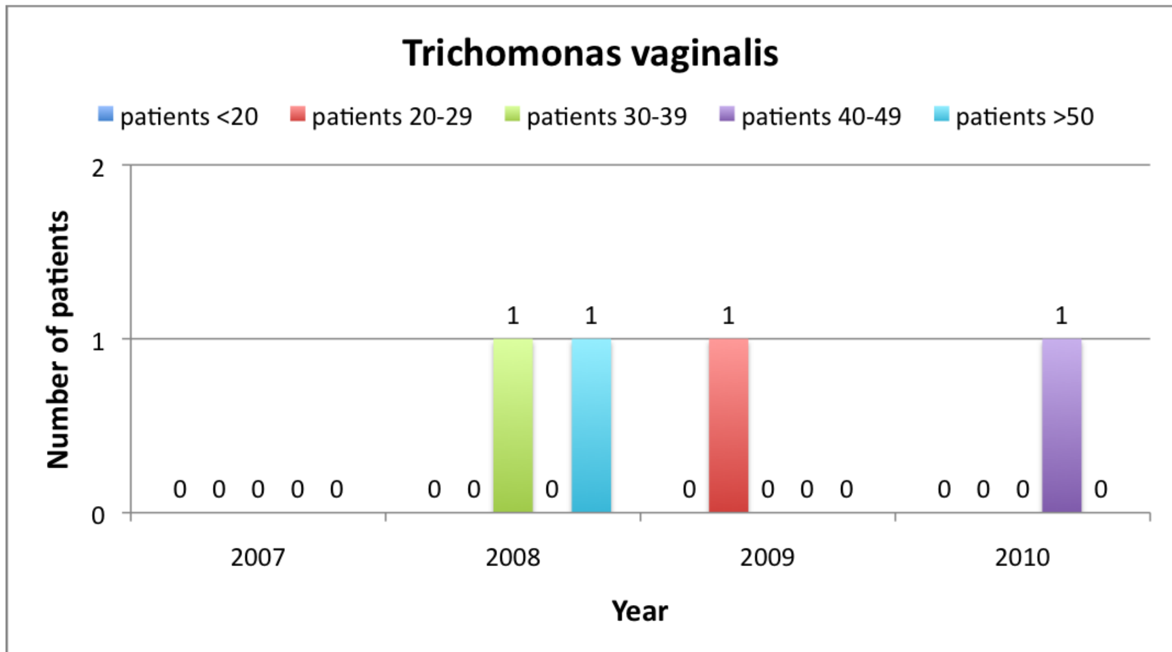


Figure 39 – Age distribution of Trichomonas vaginalis

4 Discussion

The aim of this retrospective analysis was to collect epidemiological parameters and to shed light on the distribution of the studied pathogens within the patient population in terms of age, sex and infection rates. To answer these questions, only results from examinations and smears that were taken at the STD outpatient clinic at the University Clinic for Dermatology and Venereology Graz were used. Cases that have been reported by other institutions or doctors were not included in this work.

It should be particularly emphasized on the heterogeneity of data reporting and control measures in the STI diagnostics worlwide. The problem resides within the lack of unified testing and control measures, which significantly complicate the direct comparison of numbers of cases and the evaluation of factors between individual European countries. (17)

1669 patients were tested in the period of time from May 2006 to May 2011, by the team of the STD outpatient clinic. Out of all the 1669 tested patients, 69 % (n = 1159) were of male gender and 31% (n = 510) were female patients.

The predominant proportion of men among the patients can be attributed to the fact that women often consult a gynecologist first and the STD clinic only secondarily.

The average age in male patients was at 35.7 years, in female patients at 37 years and the overall mean age at the time of testing was at 36.1 years.

Among all of the 1669 patients that underwent examination at the STD outpatient clinic, 39.4% (n= 657) were tested positive for at least one of the pathogens of interest. Out of the 657 positive tested patients, 66% (n = 431) were of male and 34% (n = 226) of female gender.

The average age in patients who tested positive for at least one of the pathogens was at 31.1 years in male patients, 33,3 years in women and the overall mean age was at 31,8 years. (table 3)

The most common pathogen found in the statistically comparable study population from 2007 to 2010 in both male and female patients was U. urealyticum (n=319), followed by C. trachomatis (n=108), N. gonorrhoeae (n=74), M. hominis (n=11), and T. vaginalis (n=4).

The STD outpatient clinic Graz was frequented both by men and women most commonly between the age of 20 and 29 years. Among men 33.8% (n = 392) and 31.1% of women (n = 159) were part of this age group. With increasing age (>30) the frequency of

consultations at the Graz STD outpatient clinic in both sexes declined, most likely due to the higher sexual activity in young adulthood and thus increased incidence of sexually transmitted infections.
However, contrary to the main trend, the STD outpatient clinic was visited by more patients over 50 (22.7%) years, than patients under the age of 20 (4.7%).
The overall trend continued in the group of positive tested patients, except for the fact that the group of < 20 year olds and the ≥ 50 year olds aligned and both amounted for 9.6% of patients. (table 3)

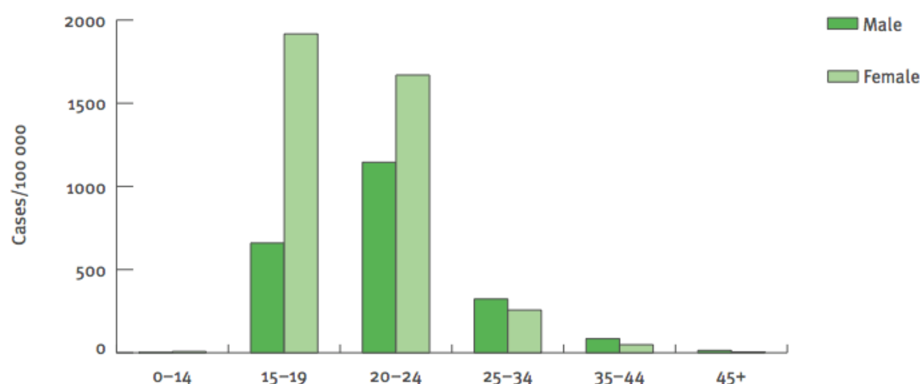
With almost 320 000 cases reported in 2010, Chlamydia infection is the most common STD in the European Union. The reporting rates have more than doubled in the last 10 years, but seem to have reached a plateau in 2010. (96)
C. trachomatis was detected in 94 male patients (27.8% of all men) and in 14 female patients (7.9% of all women) in our study population from 2007 to 2010. The distribution of cases by sex remained unchanged over the years.
The percentage of reported positive tests remained at a similar level over the studied period of 4 years. So far there is no national coverage of C. trachomatis cases in Austria, but according to data of the European Centre for Disease Prevention and Control (ECDC) there were 822 cases in 2007, 742 cases in 2008, 597 cases in 2009 and 1085 cases in 2010 detected and reported by the AT – STISentinella (table 7) regarding Austria. (96)
The AT – STISentinella is an active voluntary case based sentinel program without national coverage, which collects and uploads data to the European Surveillance System (TESSy). The STD outpatient clinic Graz is not part of the AT – STISentinella program. In comparison to AT – STISentinella data, the positive tested patients of our study would account for 3.3% of all confirmed C. trachomatis infections in Austria in 2007, 3.9% in 2008, 4.2% in 2009 and 2.5% in 2010. With a total of 108 cases within the period of four years, the catchment area of the Graz STD outpatient clinic represents an area of relativly low prevalence. The distribution of C. trachomatis within the countries of the EU varies from below 1 to more than 600 cases per 100.000 people, which is partly due to the measures taken by Member States to improve the diagnosis and reporting of infections, including active case finding measures. This being the reason, that countries with national coverage of C. trachomatis infections (United Kingdom, Netherlands and especially the Scandinavian countries) reported higher rates of infection than the central or eastern parts of the EU. (96) The prevalence for C. trachomatis in the WHO European region lies at 3.9% in women an 3.8% in men. (97)

	Number of cases			
	2007	**2008**	**2009**	**2010**
STD – outpatient clinic Graz	27	29	25	27
Austria (ECDC)	822	742	597	1085
Luxembourg	-	2	0	2
Cyprus	0	1	4	3
Malta	72	106	61	138
Slovakia	91	117	227	184
Slovenia	201	127	130	176
Turkey	205	163	103	-
Spain	218	-	845	947
Lithuania	403	403	326	367
Poland	627	-	908	539
Hungary	699	754	711	710
Latvia	711	704	1127	1042
Estonia	2480	2200	2015	1737
Belgium	2480	2601	2988	3314
Switzerland	5336	6111	6280	6575
Netherlands	7801	9436	9779	11530
Finland	13973	13871	13317	12814
Denmark	25795	-	-	-
Ukraine	35079	36584	-	-
Sweden	47101	42001	37780	36814
United Kingdom	123629	203475	214227	215501
Europe	256888	338448	346054	345421
USA	1.108.374	1.210.523	1.244.180	1.307.893

Table 6 – Number of reported confirmed Chlamydia infection cases in EU/EEA countries, 2007–10 (96,98,99)

The distribution of cases by sex and age group has not changed in the examined years from 2007 to 2010 within the study. 47.2% of cases were in the age group of 20 – 29 year-olds and 24.1% in the age group of 30-39 year-olds. Only 5.5 % of the cases involved patients over the age of 50 years. (table 5)

The age group of less than 20 years olds accounted for only 13% of all chlamydial infections and was therefore underrepresented compared to EU (ECDC) published high rates of Chlamydia infections within that age groupe. (fig. 40)



Source: Country reports from Belgium, Cyprus, Denmark, Estonia, Finland, Iceland, Latvia, Lithuania, Luxembourg, Malta, Norway, Romania, Slovakia, Slovenia, Sweden, United Kingdom.

Figure 40 – Rates of reported confirmed Chlamydia infection cases, by age and gender, in EU/EEA countries, 2010 (96)

In summary, Chlamydia trachomatis infections can be held responsible for a whole range of serious diseases in women and men, which could be at least partially prevented by early diagnosis and therapy. Table 8 represents the possible complications of untreated chlamydial infections. Without treatment, chlamydia infection of women can spread and cause abdominal inflammation as well as an agglutination of the fallopian tubes due to the inflammation, which may cause infertility. The risk of an ectopic pregnancy and preterm is increased.

In men the inflammation of the epididymis and prostate are causes for infertility.

Reiter's disease is a very rare complication, which is characterized by joint pain, urinary tract infections, eye infections and rashes expressed on the mucous membranes and skin. It affects mostly young men.

<table>
<tr><th></th><th>Clinical manifestations</th><th>Other Information</th></tr>
<tr><td>Male</td><td>urethritis, epididymitis, orchitis, infertility</td><td rowspan="3">It is estimated that 85% of women and 40% of men are asymptomatic. If untreated, chlamydial infection may persist for years. It is estimated that up to 40% of women with an untreated infection develop PID and that one in four women with PID will develop infertility. (12)</td></tr>
<tr><td>Female</td><td>cervicitis, endometritis, salpingitis, PID, infertility, preterm rupture of membranes during pregnancy, perihepatitis</td></tr>
<tr><td>Both sexes</td><td>proctitis, pharyngitis, Reiter's syndrome</td></tr>
</table>

Table 7 – Clinical manifestations of chlamydial infection (97)

N. gonorrhoeae is one of four pathogens that have to be reported to the competent authorities, as regulated by law (“Geschlechtskranheitengesetz”). (2) In our study population from 2007 to 2010 it was significantly more often detected in male patients (n=66/ 19.5% of all men) than in female patients (n=8/ 4.5% of all women). This proportion remained about the same over the years.
Every year the Austrian Federal Ministry of Health (BMG) publishes data on the reported infections for all federal states and the whole of Austria. (100) (table 9)
The positive tested patients of this study would account for 3.0% of all confirmed N. gonorrhoeae infections in Austria in 2007, 2.3% in 2008, 3.1% in 2009 and 1.7% in 2010 and. Data collected by other institutions such as the ECDC or the Centralized Information System for Infectious Diseases (CISID) of the WHO report different numbers of infections. The discrepancy may be caused by different collection systems of data. The ECDC receives its gonorrhea infection data the same way as for C. trachomatis from the AT – STISentinella. The CISID WHO/Europe infectious disease data set is compiled from reports submitted by member states (Austria/BMG). Therefore it is surprising that the data of the BMG and CISID still vary in three out of four years.
In addition also different numbers of cases and trends in the various federal states can be noticed.
The N. gonorrhoeae infections data from 2007 - 2010 for Styria was especially interesting. While we detected 19 cases in our patient population in 2007, the whole of Styria only accounted for 15 cases in the BMG report, despite the fact, that cases of the STD outpatient clinic had been reported to the authorities. This mismatch continued over the following years (19 cases in our study population/ 12 in the whole of sytria in 2008, 17/5 in 2009, and 19/4 in 2010), and therefore leads to the conclusion that the reporting system for infections needs to be revised and expanded in Styria.
Compared to the other 8 federal states the infection rates or their detection are significantly lower. While having the fourth biggest population in Austria, Styria is only at 7th place in absolute numbers of N. gonorrhoeae cases.
In the 2012 ECDC surveillance report Austria alongside France and Greece was the only participating member state without a national STI coverage system. However, according to the report Austria showed similarly high infection levels as Sweden or Switzerland and was to be found in the top third in Europe.
Regarding gonorrhea infections the highest numbers were recorded by far in the USA, Russia, the UK and Ukraine. It is remarkable that incidence rates in Austria as well as

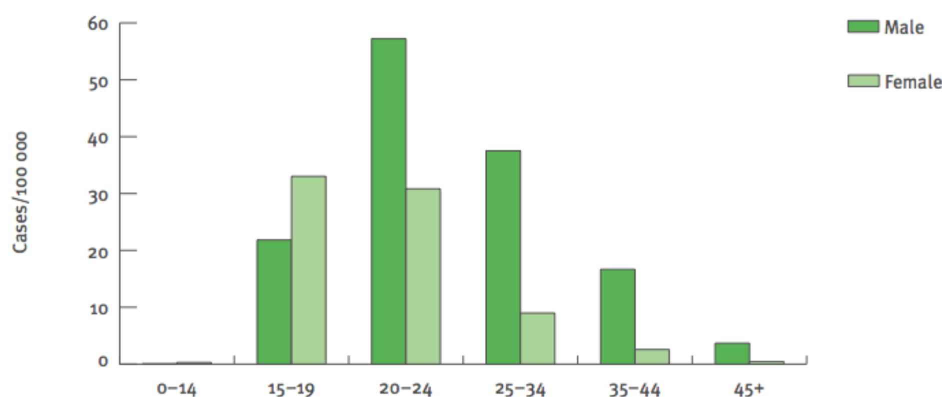
other countries (Switzerland, Finnland, Sweden, Hungary, Netherlands) are engaged in an upwards trend, however infection rates in Graz remained stable. The prevalence for N. gonorrhoeae in the WHO European region lies at 0.3% in women an 0.2% in men. (97)

	Number of cases			
	2007	**2008**	**2009**	**2010**
STD – outpatient clinic Graz	19	19	17	19
Austria (ECDC) (96)	131	263	143	339
Austria (CISID) (98)	633	821	542	1100
Austria (BMG) (100)	636	825	542	1138
Styria	15	12	5	4
Vienna	511	707	451	1015
Carinthia	11	17	9	7
Lower Austria	19	13	3	10
Upper Austria	32	26	41	55
Salzburg	22	30	21	32
Vorarlberg	1	1	3	2
Burgenland	0	0	1	0
Luxembourg	1	9	6	3
Cyprus	5	2	7	23
Slovenia	42	43	30	44
Malta	53	49	63	47
Slovakia	101	157	169	126
Estonia	174	146	127	109
Finland	193	201	238	256
Poland	330	-	402	301
Denmark	351	396	562	481
Spain	471	-	751	918
Lithuania	471	533	391	315
Turkey	518	493	152	-
Belgium	585	728	705	782
Sweden	642	725	-	842
Latvia	669	487	433	357
Switzerland	871	905	897	1173
Hungary	1041	892	872	1170
Netherlands	1827	1964	2424	2816
Ukraine	13841	12537	-	10151
United Kingdom	18631	16451	17409	18580
Europe	30063	27972	29415	31554
Russian Federation	85979	79718	67633	60058
USA (CDC)	355.991	336.742	301.174	309.341

Table 8 – Number of reported gonorrhoea cases in EU/EEA countries, 2007–10 (98,99)

The positive tested patients of our study were in accordance to EU (ECDC) published age distribution data. (96) (fig. 41)

50% of both men and women in the study who had been infected with gonorrhea were younger than 30 years at diagnosis. Most cases were found in the age group of 20 – 29 year-olds, again followed by the group of 30 to 39 year-olds. Less than 10% of cases occurred in people over 50 years. (table 5)



Source: Country reports from Belgium, Czech Republic, Denmark, Estonia, Finland, Greece, Iceland, Latvia, Lithuania, Luxembourg, Malta, Norway, Portugal, Romania, Slovakia, Slovenia, Sweden, United Kingdom.

Figure 41 – Rates of reported confirmed gonorrhoea infection cases, by age and gender, EU/EEA countries, 2010 (96)

As with Chlamydia trachomatis an ascending infection with N. gonorrhoeae can lead to severe consequences. (table 11).

	Clinical manifestations	**Other information**
Male	urethritis, epididymitis, orchitis, infertility	It is estimated that 30–80% of women and 5% of men with genital gonorrhea are asymptomatic. It is estimated that up to 40% of women with an untreated infection will develop pelvic inflammatory disease (PID) and that one in four women with PID will develop infertility. (12)
Female	cervicitis, endometritis, salpingitis, PID, infertility, preterm rupture of membranes during pregnancy, perihepatitis	
Both sexes	Conjunctivitis, corneal scarring and blindness	

Table 9 – Clinical manifestations of Neisseria gonorrhoeae infection (97)

U. urealyticum was the most wide spread pathogen in the tested patient population. U. urealyticum could be found in 173 male patients (51.2% of all men) and 146 female patients (82.0% of all women).
The number of detected positive test results of U. urealyticum declined steadily from a peak in 2007 (n=105) until 2010 (n=58). The decrease was observed both in men and women, with an accompanying shift from more positive tested males to females, from a starting 46.7% female in 2007 to 48.2% female patients in 2010. When looking at the picture of age distribution, it becomes visible, that it follows the overall trend. Patients between the ages of 20-29 years accounted for 41.4% by the age groups of 30-39 years (21.9%), 40-49 years (15.4%), ≥ 50 years (11.9%) and last by the < 20 years (9.4%). (table 5)
The detection of M. hominis was comparably low to the other microorganisms except U. urealyticum.
M. hominis was detected in 4 male patients (1.2% of all men) and in 7 female patients (3.9% of all women) in our study population. The age group of 20-29 year olds represented for 45.4% (n= 5) out of all 11 positive tested patients. Followed by the age groups of 40-49 years (n= 3; 27.3%), < 20 years (n= 2; 18.2%), and last by the ≥ 50 years (n= 1; 9.1%).
In contrast to the general age distribution of all positive tested patients there are no cases of patients between the ages of 30-39 over the whole examined time spectrum. (Table 4)
Genital mycoplasmas can be problematic in certain cases. U. urealyticum may cause an infection (NGNCU) the first time the urethra is affected.
The role of M. hominis and U. urealyticum in prostatitis, epididymitis, SARA as well as in infertility is still little studied and not yet determined.

T. vaginalis was only detected 4 times (0.8% of all positive cases) throughout the whole study population from 2007 to 2010. Out of these four cases only one male patient was infected, making T. vaginalis the pathogen with the highest percentage (75%) of female patients.
Studies conducted in developing countries (table 11) show significantly higher infection rates than this study or comparable studies of western countries. For example Fonck et al. described a prevalence of 23.0% of trichomonas infections in Kenyan women.
Owing to the small number of positive cases in our study population, a meaningfull statement on age distribution is not possible.

Study	Year	Country	Age	Study size	Diagnostic method	Prevalence	
						Male	Female
STD – outpatient clinic Graz	2007-2010	Austria	16-81	n= 516 (m:338; f:178)	Urethral swab, urine: culture and PCR	0.3%	1.7%
Price et al. (102)	2004	Malawi	18-63	n= 756	Urethral swab, urine: culture and PCR	19.8%	23%
Rassjo et al. (103)	2006	Uganda	-	n= 306	Vaginal swab, urine: PCR	0.0%	8.0%
Fonck et al. (104)	2000	Kenya	15-52	n= 621	Vaginal swab: microscopy	-	23.0%
Leutscher et al. (105)	2005	Madagascar	15-49	n= 643	Vaginal swab (females), urethral swab (males): culture	7.4%	23.4%
Klinger et al. (106)	2006	Tanzania	20-44	n= 2028	Urine: PCR	6.3%	10.7%
Buve et al. (107)	2001	Sub-Saharan Africa	15-49	n= 8000	Vaginal swab: culture	-	3.2% - 34.3%
Bowden et al. (108)	1999	northern Australia	12-73	n= 1090	Tampon: PCR	-	25.0%
Miller et al. (109)	2005	USA	> 18	n= 12449	Urine: PCR	1.7%	2.8%
Joyner et al. (110)	2000	USA	-	n= 454	Urine: Culture	2.8%	-
Lo M. (111)	2002	New Zealand	-	n= 88	-	-	2.0%
Van Der Pol et al. (112)	2005	USA	14-17	n= 268	Vaginal swab: PCR		6.0%

Table 10 – Prevalence of Trichomonas vaginalis (113)

Trichomoniasis, a potential risk factor responsible for a number of complications (table 12), including the risk of miscarriage, premature birth in pregnant women or decreased birth weight of newborns, still remains mostly disregarded.

	Clinical manifestations	Other information
Male	urethral discharge (NGNCU) often asymptomatic	It is estimated that up to 80% of women with laboratory confirmed T. vaginalis infections are asymptomatic. (114)
Female	vaginosis with profuse, frothy vaginal discharge; preterm birth, low birth weight babies	

Table 11 – Clinical manifestations of trichomoniasis (97)

Further research, cheaper diagnostic tests and screening methods are necessary considering T. vaginalis being the most common STI worldwide, with a especially high prevalence in developing countries.

In conclusion, the pathogens detected by this study are prove, that STIs are still a health problem, which often is misunderstood or underestimated. The numbers for the STD outpatient clinic Graz show that the prevalence of especially C. trachomatis and N. gonorrhoeae are still on a constant level.
We also have to be aware of the possibility that the number of asymptomatic as well as unregistered infected is considerably higher. Therefore we should reevalute and revise our detection system in order to get more infected persons treated.

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