"CAROL DAVILA" UNIVERSITY OF MEDICINE AND PHARMACY,

BUCHAREST

DOCTORAL SCHOOL

FIELD OF MEDICINE

GENITAL INFECTION WITH UREAPLASMA AND MYCOPLASMA

– INCIDENCE AND ANTIBIOTIC RESISTANCE

PhD THESIS SUMMARY

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Content

The fundamental problem

Sexually transmitted diseases (STDs) are a major public health problem worldwide. In developing countries, STDs are among the top five reasons for seeking health services. The term STD refers to the various clinical symptoms caused by pathogenic microorganisms that are transmitted through sexual contact. Among the causative agents of STDs are Ureaplasma and Mycoplasma species, whose role in sexually transmitted infections remains hotly debated [1],[2],[3],[4].

Data from the specialized literature indicate that the prevalence of Ureaplasma and Mycoplasma species is ~21% and ~3%, respectively, and the prevalence of Chlamydia trachomatis is also ~3% [3],[4].

The majority of infections with Ureaplasma, Mycoplasma and/or Chlamydia are completely asymptomatic (~80%). For this reason, it is essential that these microorganisms are included in STD screening, given that if left untreated they can lead to severe complications in both male and female patients [5],[6],[7].

Working hypothesis and general objectives

This work will be based on a retrospective, single-center study based on the analysis of observation sheets of patients with genital infections with Ureaplasma, Mycoplasma and Chlamydia included in the database used in the Dermatovenerology department of Ponderas Academic Hospital, during the period January 2021 - December 2021.

The objectives of this work are to evaluate the prevalence of genital infections with U. urealyticum, M. hominis and C. trachomatis in the Romanian population, in relation to the presence or absence of urogenital symptomatology, as well as to evaluate the antibiotic sensitivity and resistance of U. urealyticum and M. hominis strains to antibiotics currently available in Romania.

At the same time, this work emphasizes the importance of screening and appropriate antibiotic treatment of genital infections caused by Mollicutes. Left untreated, these infections can lead to serious complications, including infertility, a condition that has become a significant global health problem.

General Research Methodology

This study was conducted over a period of one year, from January 2021 to December 2021 in the Dermatovenerology Department of Ponderas Academic Hospital in Bucharest, Romania. Data were collected and analyzed on all patients presenting for genital infections with Ureaplasma and Mycoplasma. Samples were obtained from two groups of patients: symptomatic subjects with urogenital symptoms such as burning or pain during urination, vaginal/urethral discharge, and asymptomatic subjects who came for STD screening – many of whom reported sexual contact with infected persons. Only samples collected during the first visit were considered, excluding problems obtained during follow-up from the same patient.

The study was approved by the ethics committee of Ponderas Academic Hospital. Informed consent was obtained from all patients.

Male patients were placed in the gynecological position and asked to retract the foreskin of the penis and keep it retracted throughout the procedure. The doctor used sterile cotton or gauze to clean the opening of the urethra at the tip of the penis. To facilitate sample collection and stimulate prostatic gland secretion, prostatic massage was performed prior to collection of the sample. Then, a first cotton swab was gently inserted ~2 cm into the urethra and rotated. To obtain a good sample, the test was performed ≥3 days from the last sexual intercourse and 2 h after urination. The swabs were placed in R1 broth from a Mycoplasma IST 2 kit (bioMérieux) to initiate the isolation of mycoplasmas. For female patients, the vaginal sample was taken by placing the patient in a gyneco- logical position and carefully introducing a cotton swab into the vaginal canal. The use of commercial lubricants or antiseptics was avoided. The swabs were placed in R1 broth to initiate the isolation of Mollicutes. The liquid medium for *U. urealyticum* and *M. hominis* was a transport medium used for inoculation of a test strip. In order to perform the phenotypic identifica- tion of *U. urealyticum*, urea broth was used, which contained medium base (pleuropneumonia-like organism broth), yeast extract, horse serum and urea. To determine the growth of this microorganism, phenol red was added to the culture medium, as it changes from red to intense raspberry red in the presence of urease and ammonium production. The culture medium specific for *M. hominis* included arginine which, when metabolized, produces an alkaline compound that changes phenol red to a raspberry red color. The culture media were incubated at 37℃ until the phenol red indicator changed color. The Mycoplasma IST 2 kit was used according to the manufacturer's instructions as follows. As aforementioned, the sample-bearing swab was placed in the transport medium R1 broth (3 ml). The broth was mixed with the contents of the lyophilised

R2 vial provided with the kit, which contained the substrates necessary for the develop- ment of microorganisms. A volume of 55 ml was added to each of the 22 domes in the test strip. Firstly, the phenotypic detection of *M. hominis* and *U. urealyticum* was performed. Secondly, the microorganisms were quantified, to determine whether the sample concentration was >1x10^4 change color-changing units (CCU), as this indicated an important presence of these micro- organisms (positive result).

A second swab was inserted in the urethra of male patients and the vaginal canal of women to collect urogenital samples for the detection of *C. trachomatis* by PCR. The samples were collected using a DNA collection device, comprising a cyto- brush and DNA holder buffer (Specimen Transport Medium; Digene; Qiagen, Inc.), for the investigation of bacterial infec- tions. Bacterial DNA samples were extracted from samples collected from the urogenital tract using an RTP®-Bacteria DNA kit (Invitek Diagnostics), according to the manufacturer's procedures and amplified using a 5TD6 ACE Detection kit (Allplex STI Essential Assay; Seegene, Inc.) for the detection of *C. trachomatis* by PCR. The PCR primer sequences are not disclosed by the manufacturer. The PCR conditions were as follows: 1 cycle of 94℃ for 15 min, 40 cycles of 94℃ for 30 sec, 63℃ for 90 sec and 72℃ for 90 sec, and 1 cycle of 72℃ for 10 min. The amplification of plasmidial DNA as an internal control occurred in the same reaction. The PCR product was subjected to electrophoresis on a gel containing 2% agarose stained with ethidium bromide. Amplification of the target was only observed when the respective bacterial DNA was present in the clinical sample. This method was applied to 30 samples. For the remaining 236 the testing method was changed due to financial issues at the laboratory. The subsequent method used for the detection of *C. trachomatis* involved the insertion of a swab in the urethra for male patients and the vaginal canal for women to collect urogenital samples for analysis by an alternative PCR method. The urethral sample was transferred into an Aptima Swab Specimen Transfer Tube (Hologic, Inc.). The samples were transported at a temperature of between 2 and 30℃. Determination of *C. trachomatis* rRNA in the genital secretions was performed using the Panther® System analyzer (Hologic, Inc.), which is based on nucleic acid amplification testing with transcription-mediated amplification (TMA) and dual kinetic detection. TMA is an isothermal amplification method that uses RNA polymerase and reverse transcriptase. Since the amplification temperature is 37-42℃, the technique does not require a thermocycler and can be performed using a thermoblock. TMA uses two primers that flank the region to be amplified: A promoter primer and a non-promoter primer with the same sense as the target. The 3' end of the promoter primer is complementary to that of the target RNA and the 5' end is recognized by RNA polymerase. Amplification is initiated via the binding of the promoter primer to the target RNA, which is then reverse transcribed to generate cDNA. The DNA-RNA duplex is degraded, and the RNA released through the RNase H activity of the reverse transcriptase. The second primer binds to the cDNA and generates double-stranded molecules. Hundreds of copies of the RNA amplicons are thus transcribed by means of this DNA and each copy can be converted into new double-stranded DNA molecules. The amplification prod- ucts are analyzed by hybridization with oligonucleotide probes labeled with chemiluminescent substances. Assay results were automatically interpreted by APTIMA Assay software (Panther System®; Hologic, Inc.) using the APTIMA Combo 2 protocol, and presented as individual CT test results. Based on the kinetic type and total relative light units (RLU) in the detection step, the test results were assigned as negative (RLU <25), equivocal (RLU <100), positive (RLU >100) or invalid. The categorical variables were expressed as count and percentage. Association tests were performed by chi-square or Fisher's exact test, accordingly. The statistical significance was assumed if a null hypothesis could be rejected at a p-value of 0.05. The multiple antibiotic resistance (MAR) index was calculated as the ratio between the number of antibiotics that an isolate is resistant to and the total number of antibiotics to which an organism is exposed. MAR index values greater than 0.2 indicate that antimicrobial resistance is high. The MAR index values for *M. hominis* and/or *U. urealyticum* were compared to a non-parametric test: Kruskal-Wallis (p = 0.0003614), considering the normality conditions were not fulfilled (Shapiro- Wilk normality test, p = 0.0002859). The data were analysed using the R Statistical Software version 4.4.1.

Prevalence of Ureaplasma urealyticum, Mycoplasma hominis and Chlamydia trachomatis in symptomatic and asymptomatic patients

The study population consisted of 266 patients (225 males and 41 females) aged between 18 and 80 years, and 89% of the patients were aged 31-50 years. Among these 266 patients, 59 (22%) had STD symptoms while the other 207 patients (78%) had no STD symptoms at all. Regarding the symptomatic patients, 8 (14%) had U. urealyticum, 1 (2%) had U. urealyticum and M. hominis coinfection, 3 (5%) had C. trachomatis, and no symptomatic patients were infected with M. hominis alone (all $P<0.001$).

Symptoms of urethritis were reported by 27/266 (10%) of the patients. Of those 27 patients, 4 (15%) had U. urealyticum and 1 (4%) had C. trachomatis, but none of the patients with symptoms of urethritis had U. urealyticum and M. hominis coinfection or were infected with M. hominis alone (all $P<0.001$).

In the present study, 23/266 (9%) patients presented symptoms characteristic of prostatitis. These comprised 3 patients (13%) whose prostatitis was caused by U. urealyticum and 1 (4%) in which the prostatitis was caused by C. trachomatis, while prostatitis was caused neither by U. urealyticum and M. hominis coinfection, nor by M. hominis alone in these patients (all $P<0.001$). Symptoms of both urethritis and prostatitis were present in 6/266 (2%) of the patients. Only 1/6 patients with these symptoms (17%) was infected with C. trachomatis ($P=0.1025$; Table II). Neither of the Mollicutes was detected in patients with urethritis and prostatitis ($P=0.003892$).

The study population included 207/266 (78%) patients who were completely asymptomatic. Among these 207 patients, 29 (14%) were discovered to have U. urealyticum, 13 (6%) were coinfected with both Mollicutes, 1 (0%) was infected with M. hominis alone and 4 (2%) were infected with C. trachomatis (all $P<0.001$).

Regarding the female patients, there were only 3 cases of vaginitis, one of which was a symptomatic coinfection with Mollicutes. The remaining 38 women had asymptomatic genital infections.

The association between the Mollicutes and C. trachomatis was also evaluated. It was found that 2 patients (14%) with U. urealyticum and M. hominis coinfection were also infected with C. trachomatis, while none of the patients who were infected with U. Urealyticum or M. hominis alone were also positive for C. trachomatis ($P=0.1071$).

In conclusion, the data from this study support the need for careful monitoring of genital infections, promotion of sexual health education, and easy access to testing, so as to prevent the transmission of pathogens and reduce the long-term negative effects on reproductive health.

Antimicrobial resistance of Ureaplasma urealyticum and Mycoplasma hominis in the Romanian population

The study population consisted of 52 patients (34 males and 18 females) with U. urealyticum and/or M. hominis. 28 of the male patients (82%) and 9 of the females (50%) were diagnosed with U. urealyticum genital infection. In contrast, 5 males (15%) and 9 females (50%) were positive for both U. urealyticum and M. hominis (Figure 1). There is a statistically significant association between gender and a particular isolate ($p = 0.01914$). There was only 1 male (3%) with M. hominis.

The average age for the U. urealyticum patients was 33 for male and 25 for female patients; for those who had mixed isolates, the average age was 29 for males and 32 for females. The male patient with M. hominis was 51 years old.

Forty-three patients (27 males and 16 females) were asymptomatic, and 9 (7 males and 2 females) experienced symptoms suggestive of balanitis, prostatitis and vaginitis, respectively. None of the women included in the study was pregnant.

14 out of the 52 patients with genital infections caused by U. urealyticum and, or M. hominis tested positive for coinfection with both isolates. All of them (100%) were sensitive to doxycycline and tetracycline. In addition, 21% of the patients were susceptible to ofloxacin. However, 71% of U. urealyticum + M. hominis mixed isolates showed an intermediate pattern to ofloxacin on the antibiograms, in contrast to just 7% that were resistant to ofloxacin. Mixed isolates showed extremely low susceptibility to ciprofloxacin, erythromycin, azithromycin, and clarithromycin (7%). Erythromycin, azithromycin, and clarithromycin presented an identical pattern on the antibiograms (susceptible in 7% of the cases, intermediate in 29% of the patients, and resistant in 64% of the cases). The second most resistant antibiotic is ciprofloxacin, with a 57% rate of resistance and a 36% rate of intermediate response. A significant association was found between the resistant strains and the type of antibiotic ($p < 0.001$).

Regarding the U. urealyticum isolates, all 37 patients (100%) were susceptible to doxycycline and tetracycline. This mollicute showed 84% susceptibility to clarithromycin, followed by erythromycin and azithromycin, with a percentage of 76% and 73%, respectively.

U. urealyticum strains were the least susceptible to ciprofloxacin (11%) and ofloxacin (19%). Moreover, these strains were the most resistant to ciprofloxacin (51%) compared to ofloxacin (16%), erythromycin (8%), azithromycin (8%) and clarithromycin (8%). The intermediate response rates were ofloxacin (65%), ciprofloxacin (38%), azithromycin (19%), erythromycin (16%) and clarithromycin (8%). A significant association was found between the resistant strains and the type of antibiotic ($p < 0.001$).

Out of the total of 52 patients included in the study, there was just 1 patient that tested positive for M. hominis genital isolate. The antibiogram exposed that the isolate was susceptible to doxycycline, tetracycline, ciprofloxacin and ofloxacin and resistant to erythromycin, azithromycin and clarithromycin.

Antibiotic treatment lasted an average of 30 days for all individuals. Regarding U. urealyticum, in 9 patients (24%) the infection was eradicated, in 4 (11%) it remained persistent, and in 24 (65%) cases the patients did not present themselves for post-treatment control. In the case of the patient with M. hominis it is not known whether the infection was eradicated or not. For coinfection with Mollicutes, in 5 patients (36%) the infection was eradicated, and 9 (64%) did not present themselves for control. No case of persistence of coinfection was documented. Regarding the eradication of infection by gender and Mollicutes species, in 8 men the infection with U. urealyticum was eradicated and in 2 the coinfection. In the case of women, one patient with U. urealyticum and 3 patients with double infection with Mollicutes were found negative at the control examination. Of the patients who did not attend for post-treatment follow-up, 18 males had U. urealyticum infection, 1 male had M. hominis infection, and 3 had dual infection. An equal number of females (6) had both U. urealyticum infection and coinfection. Persistence of infection was reported in 2 males and 2 females for solitary U. urealyticum infection.

The multiple antibiotic resistance (MAR) index has been calculated. The result was 0.33 for U. urealyticum isolates, 0.64 for genital coinfection with both U. urealyticum and M. hominis, and 0.42 for M. hominis isolates. The differences between the MAR index values in relation to the isolates are particularly statistically significant ($p = 0.0003614$).

In conclusion, the data obtained underline the importance of systematic testing, rational use of antibiotics and prevention of the development of bacterial resistance. Only through a multidisciplinary approach and a responsible attitude towards antimicrobial therapy, we can improve the management of genital infections and contribute to reducing the impact of microbial resistance on public health.

An overview of the relationship between Mollicutes, infertility, and antibiotic resistance

This study was a literature review, collecting and comparing the results of recent studies in the field from the Pubmed database. The keywords used for searching within the database were: "Ureaplasma", "Ureaplasma urealyticum", "Ureaplasma parvum", "Mycoplasma", "Mycoplasma hominis", "Mycoplasma genitalium", "Mollicutes", "infertility caused by Ureaplasma", "infertility caused by Ureaplasma urealyticum", "infertility caused by Mycoplasma", "infertility caused by Mycoplasma hominis", "infection with Ureaplasma in men", "infection with Mycoplasma in men", "infection with Ureaplasma in women", "infection with Mycoplasma in women".

Infertility is a complex condition, determined by a combination of genetic, microbiological and environmental factors, affecting both women and men. Among microbiological causes, bacteria of the class Mollicutes – particularly Ureaplasma and Mycoplasma – play a significant role in reproductive health. These microorganisms have been extensively studied for their ability to disrupt normal reproductive processes and induce genetic and epigenetic changes that may contribute to infertility [8],[9],[10].

Identification of Mollicutes infections in infertile couples is not routinely included in standard infertility testing protocols. This is mainly due to the limited scientific evidence linking these microorganisms to infertility, but also to the lack of awareness among clinicians about the impact of Ureaplasma and Mycoplasma infections on reproductive health [10],[11],[12]. However, recent studies suggest that these bacteria may play a significant role in reducing fertility, affecting both men and women. Therefore, increasing awareness among specialists and expanding screening for these infections could contribute to improving the clinical management of patients with conception problems [9],[13],[14].

Mollicutes infections not only affect the integrity of the reproductive tract but can also cause genetic and epigenetic changes that negatively influence fertility. These microorganisms have been associated with sperm DNA fragmentation, impaired oocyte quality, and reduced endometrial response, which can compromise the success of conception. The presence of these bacteria can also generate chronic inflammation, which, over time, can lead to impaired reproductive function. Therefore, the integration of molecular tests into diagnostic routines could improve early identification of infections, thus preventing their negative effects on fertility [11],[14],[15].

Male and female fertility are fundamental factors in human reproduction, and a detailed understanding of the biological and pathogenetic mechanisms involved is crucial for the development of effective diagnostic and treatment strategies. In this context, future research should focus on elucidating the mechanisms by which Mollicutes affect reproductive function and on the development of personalized therapeutic interventions. Early diagnosis and personalized therapeutic approaches can significantly increase the chances of success for couples facing infertility, thus contributing to the optimization of treatments and increasing conception rates.

Conclusions and personal contributions

This paper provides valuable insight into the prevalence, antimicrobial resistance, and clinical implications associated with U. urealyticum and M. hominis infections. These studies highlight the significant presence of these Mollicutes species in both symptomatic and asymptomatic patients, highlighting their major role in genitourinary infections. These data indicate the need for regular screening programs, especially among high-risk populations, for early detection and management of infections.

This topic (chapter 5) is of particular importance, being remarkable for its in-depth approach to the prevalence and incidence of genital Mollicutes infections. As far as we are concerned, this study represents the only research in Romania that includes both female and male patients from a major venereology center. Due to its comprehensive nature, this paper represents a valuable contribution to the specialized literature in the field.

Regional and national differences in the prevalence of genital Mollicutes infections reflect variations in medical practices, sexual behaviors, and access to health services. The study highlighted the prevalence of these microorganisms among symptomatic and asymptomatic patients, revealing significant variability between different populations. In developed countries, the prevalence of these infections is influenced by the frequent use of contraceptive methods and screening programs for sexually transmitted diseases. For example, in Western Europe and North America, the detection rate is higher, which leads to early treatment and reduced transmission. In contrast, in regions with limited access to health services, such as Sub-Saharan Africa or parts of Southeast Asia, infections often remain undiagnosed, increasing the risk of complications such as infertility or pelvic inflammatory disease. Cultural and religious differences also influence awareness and preventive behaviors, including condom use. The study also highlighted that the prevalence of C. trachomatis is frequently associated with Mollicutes infections, indicating the need for an integrated management of these infections. Thus, the study highlights the importance of regionally tailored public health strategies that include sexual education, regular testing, and affordable treatment to reduce the impact of these infections on global reproductive health.

A key aspect to highlight is the laboratory method used to diagnose infections caused by different Mollicutes species. Currently, there is no universally standardized method for their detection, with each country adopting its own techniques, depending on the availability of resources and costs. This methodological diversity can lead to diagnostic errors, which in turn lead to misinterpretation of the results. Consequently, certain strains may be misidentified, which prevents the clear establishment of their pathogenic or commensal nature.

Attention is drawn to the alarming rate of antimicrobial resistance observed in the Romanian population. The research highlights the difficulties in treating infections caused by U. urealyticum and M. hominis due to the increased resistance to commonly used antibiotics, such as tetracyclines and macrolides. This trend may not only complicate therapeutic strategies, but also requires the urgent development of rational antibiotic use management programs to counter this emerging threat. Although in this work (chapter 6) all individuals analyzed showed complete susceptibility to doxycycline, the specialized literature indicates an alarming global trend of resistance development to this antibiotic. This resistance is mainly attributed to the tet(M) gene mutation, as well as the extensive use of doxycycline as an empirical treatment for other infectious pathologies. In many developed countries, effective therapeutic alternatives have already been identified, which allows for more flexible management of resistant infections. However, in the local context, the lack of backup therapeutic options represents a major vulnerability.

If the phenomenon of doxycycline resistance were to become prevalent in our country, the management of infections caused by Ureaplasma and Mycoplasma could become a severe public health problem, with significant implications for treatments and the costs associated with medical care. It is therefore imperative to implement measures to monitor and limit the irrational use of this antibiotic, as well as to invest in access to modern therapeutic alternatives. It is essential to implement national and regional systems for monitoring the evolution of antibiotic resistance in Mollicutes bacteria, which will facilitate the updating of therapeutic guidelines. Future research should focus on discovering new antibiotics or alternative therapies, such as the use of bacteriophages or immunomodulatory treatments. Understanding the genetic and epigenetic mechanisms by which Ureaplasma and Mycoplasma develop resistance could help identify novel therapeutic targets. Further research is needed to elucidate the precise mechanisms by which Mollicutes infections influence fertility, as well as the potential benefits of preventive treatment in reducing associated infertility. By integrating these research directions and applying multidisciplinary strategies, the medical community can more effectively respond to the clinical and public health challenges posed by genital Mollicutes infections.

The review published in 2024 (chapter 7) aims to explore the broader implications of Mollicutes infections, particularly their link to infertility and antibiotic resistance. This comprehensive review highlights the negative impact of these microorganisms on reproductive health and the urgent need to develop innovative therapies and preventive measures to reduce their effects. Mollicutes infections, including Ureaplasma and Mycoplasma, have a significant impact on reproductive health, being frequently associated with infertility in both women and men. At the molecular level, these microorganisms are distinguished by their genetic adaptability, having small genomes and an increased capacity for antigenic variation, which allows them to evade the host immune response and develop antibiotic resistance. In addition, enzymes such as proteases and hydrolases produced by Mollicutes contribute to the degradation of host tissue and the perpetuation of chronic inflammation. From a genetic perspective, host polymorphisms can influence susceptibility to infections and response to treatment, paving the way for personalized therapies. Modern genomic sequencing techniques also provide insights into the complex host-pathogen relationship and the identification of molecular targets for new antibiotics or immunotherapies.

Genomic analyses have revealed the presence of genes involved in bacterial adhesion to epithelial cells, such as genes encoding surface proteins (P1, P30) and variable lipoproteins (VLPS), which allow evasion of the host immune system. A central mechanism involves the formation of bacterial biofilms, which provide protection against the immune response and antibiotics, thus increasing the persistence of infections. Transcriptomic studies have demonstrated that heat shock proteins (Hsp70) and antioxidant enzymes produced by Mollicutes modulate the host inflammatory response. This chronic inflammatory interaction can induce endometrial damage, altered tubal patency and impaired spermatogenesis in men. In addition to direct inflammatory effects, epigenetic research indicates that Mollicutes can influence host gene expression through abnormal DNA methylation and histone modifications, thus disrupting developmental processes essential for fertility. Mollicutes also produce toxins (such as vacuolating cytotoxin), which damage the DNA integrity of host cells and promote apoptosis. Another relevant aspect is the impact on the genital microbiome. The dysregulation of the microbiota, induced by Mollicutes, increases susceptibility to other infections and impairs the body's ability to maintain an optimal reproductive environment. In men, proteins secreted by these bacteria interact with sperm, reducing the mobility and integrity of cell membranes. Understanding the genetic and molecular mechanisms involved in the pathogenesis of Mollicutes opens new perspectives for the development of targeted therapeutic strategies. The identification of specific genetic markers and inflammatory pathways could improve early diagnosis and personalized treatment of infertility associated with these microorganisms.

Studies on Mollicutes highlight the need for an integrated approach, including early diagnosis, monitoring of antibiotic resistance, and development of preventive strategies such as vaccination. In the context of infertility, continued research is essential to better understand the pathogenic mechanisms and reduce the burden of these infections on overall reproductive health. In the future, a comprehensive, prospective, case-control study that takes into account different strains of Ureaplasma and Mycoplasma, the presence of other microorganisms, the level of PMN elastase as a marker of inflammation, and the number of sexual partners will be critical to confirm or refute the role of Mollicutes in infertility.

In conclusion, genital infections with Mollicutes represent an important public health problem, due to its high prevalence and increasing resistance to antibiotics. It is crucial to implement surveillance programs and optimize therapeutic guidelines to combat these challenges. In addition, standardizing detection methods, researching resistance mechanisms and developing new therapies are priorities for the future. Through a multidisciplinary approach, the impact of these infections on individual and collective health can be reduced.

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Publications

1. **Cutoiu A**, Boda D. Prevalence of *Ureaplasma urealyticum*, *Mycoplasma hominis* and *Chlamydia trachomatis* in symptomatic and asymptomatic patients. Biomed Rep. 2023 Sep 1;19(4):74. (chapter 5)

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2. **Cutoiu A** and Boda D: Antimicrobial resistance of Ureaplasma urealyticum and Mycoplasma hominis in the Romanian population. Farmacia 71: 1, 2023. (chapter 6)

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   https://farmaciajournal.com/issue-articles/antimicrobial-resistance-of-ureaplasma-urealyticum-and-mycoplasma-hominis-in-the-romanian-population/

3. **Cutoiu A**, Boda D. An overview regarding the relationship between Mollicutes, infertility and antibiotic resistance (Review). Biomed Rep. 2024 Jun 17;21(2):119. (chapter 7)

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