Pakistan J. Zool., pp 1-13, 2025. **DOI**: https://dx.doi.org/10.17582/journal.pjz/20240909140355

Review Article

Influence of Vaginal Pathogens on Female Infertility: Therapeutic Potential of Lactic Acid Bacteria in Global Public Health

S. Rehan Ahmad[1]*, Izhar Husain[2] and Pritha Ghosh[3]

[1]*Hiralal Mazumdar Memorial College for Women, West Bengal State University, Kolkata, West Bengal, India*
[2]*Department of Public Health, College of Applied Medical Sciences, Khamis Mushait Campus, King Khalid University (KKU), Abha 62561, Saudi Arabia*
[3]*National Institute of Nutrition-ICMR, Hyderabad, India*

Article Information
Received 09 September 2024
Revised 15 October 2024
Accepted 30 October 2024
Available online 08 May 2025 (early access)

Authors' Contribution
Conceptualization, data curation, writing – original draft preparation: SRA, PG
Formal analysis, investigation, methodology, resources, validation, visualization, project administration: SRA, IH, PG
Funding acquisition: IH
Supervision: SRA
Writing – review & editing: SRA, IH

Key words
Bacterial vaginosis, Female infertility, *Mycoplasma genitalium*, Pelvic inflammatory diseases, Vaginal *Lactobacillus*, Vaginal microbiota dysbiosis

ABSTRACT

Maintaining the balance of the vaginal microbiota is very much crucial for normal vaginal function and reproductive health. Both pathogenic and nonpathogenic microbes are present inside the vagina. Among them, *Lactobacillus* is the most predominant. *Lactobacillus* gives protection against a wide range of pathogenic infections by producing lactic acid, hydrogen peroxide, and bacteriocins. A low amount of *Lactobacillus* strains and a higher amount of facultative anaerobic pathogens inside the vagina lead to vaginal microbiota dysbalance. Female vaginal with a lower concentration of vaginal *Lactobacillus* are more prone to upper genital tract infections, sexually transmitted infections, and other anaerobic pathogenic infections. Dysbiosis of the vaginal microbiota is strongly associated with infertility, poor pregnancy rate, pregnancy complications, spontaneous abortion, preterm birth, and frequent abortion. Female infertility is one of the most complex reproductive diseases, and there are no effective ways to get out of this problem. Several infection conditions like bacterial vaginosis, pelvic inflammatory diseases, and endometritis are related to adverse reproductive outcomes and infertility via disturbing normal immunity, typical vaginal microbial composition, regulating pathophysiological pathways, and inducing inflammation. Pathogenic bacteria, including *Neisseria gonorrhoeae*, *Chlamydia trachomatis*, *Trichomonas vaginalis*, herpes simplex, and *Mycoplasma genitalium*, are mainly responsible for the infertile condition. The aim of this review is to show the link between vaginal microbial disbalance and female infertility. This review also summarizes the effects of various inflammatory conditions and infectious diseases of the reproductive system on female infertility. Several pathogenic microbes, including sexually transmitted microorganisms and their impacts on female infertility, are also reviewed in this paper.

INTRODUCTION

Recently, female infertility has been a serious health issue around the whole world (Tomaiuolo *et al*., 2020). Infertility, a complicated medical condition, can affect an infertile individual's physical, mental, and psychological health. Both female and male infertility can cause variofertility-related conditions. Though male fertility is a serious and prevalent factor in infertility studies this review paper only summarizes different bacterial infection-related infertility conditions in women (Walker and Tobler, 2022). According to the World Health Organization (WHO), infertility is defined as failure to achieve pregnancy after 12 months or more of regular unprotected sexual intercourse. At the same time unable to get pregnant after the very first successful pregnancy is called secondary infertility. It may present in both male and female individuals, affecting a million people worldwide. WHO reported that 186 million people and 48 million couples suffer from infertility (WHO, 2020). In the United States, nearly 6% of married women in the reproductive age group (15 to 44 years) currently have infertility and 12 % of the total married women population faces impaired fecundity problems and other pregnancy difficulties. In the United States, the male factor is one

* Corresponding author: professor.rehaan@gmail.com
0030-9923/2025/0001-0001 $ 9.00/0

of the main reasons behind infertility. 8% of infertility is because of male partners only over there (CDCP, 2021a). In India, a wide range of lifestyle diversity like working patterns, traditions, hygiene patterns, health care facilities, customs, traditions, and external environmental conditions leads to different infertility rates within the different regions and groups of people. Recently, in India, infertility is between 10 and 14%. The burden is higher in urban areas (approximately 1 in 6 couples suffer from infertility). Infertile women face psychological, social, and physical ignorance and trauma daily of the phenomenon. These mental and physical issues are more severe in India within patriarchal societies. Female infertility can put women into marital insecurity, and the consequences can exert massive emotional instability for women. Women used to face lots of disrespect, rejection, teasing, and abusive words at home and in society. Centres for Disease Control and Prevention (CDCP) have started a National Public Health Action Plan to prevent, manage and detect infertility. This plan covers some essential points regarding this infertility field, like promoting healthy behaviours to control infertility rate, early diagnosis, preventive measures, and therapeutic remedies to control infertility, and lastly, avoiding external exposures including infectious pathogens, environment, hygiene, and iatrogenic agents to secure fertility possibility (Kalidasan *et al*., 2020).

Vagina is an appropriate place for microbial growth due to its humid and warm environment and the presence of nutritional sources. 9% of the total microbiota inside the human body is occupied by vaginal microbiota. *Lactobacillus* has been found abundantly in the vaginal microbiota, and along with *Lactobacillus*, many other bacterial species such as *Bifidobacterium*, *Prevotella*, *Gardnerella*, *Megasphaera*, *Atopobium*, *Anaerococcus*, *Sneathia* are also found (Amabebe and Anumba, 2018; Sirota *et al*., 2014). Normal vaginal microbes can prevent pathogenic growth by using their anti-microbial and anti-inflammatory compounds. *Lactobacillus* use to produce L-lactic acid and D- lactic acid that maintains the normal vaginal acidic pH (4.5) (Tomaiuolo *et al*., 2020). The normal vaginal pH of a healthy female is near about 4.5. However, dysbiosis of the vaginal microbiota may increase the pH value inside the vagina, which is directly associated with a higher risk of preterm birth and female infertility. One descriptive study by Lykke *et al*. (2021) detected an elevation of the pH value in the lower vagina of the female with abnormal vaginal microbiota than the women with normal vaginal microbiota by selecting both pregnant and nonpregnant women for this study. This study also identified some pathogens like *Atopobium vaginae*, *Leptotrichia amnionii*, *Sneathia sanguinegens*, bacterial vaginosis-causing bacteria, *Prevotella* spp., TM7 among women with abnormal vaginal microbiota by using PCR technique (Lykke *et al*., 2021). *Lactobacillus* also produces bactericidal peptides, bacteriocins which can form pores on the pathogen's cell membrane and consequently rupture it. The balance between normal vaginal microbes and unwanted facultative pathogens in the vaginal microbiome plays an essential role in women's reproductive health, including fertility chances. Disbalance in the vaginal microbiota can damage the first-line defense mechanisms against pathogens. Bacterial vaginosis, an excess number of facultative anaerobes inside the vaginal microbiota instead of *Lactobacillus*, urinary tract infections, sexually transmitted infections, and preterm birth are some common reasons for vaginal microbial dysbiosis (Tomaiuolo *et al*., 2020). Infertility-causing pathogenic microbes used to enter the upper genital tract through the vagina. The vaginal microbiota is composed of many anaerobic as well as aerobic bacteria; any external factors, including medications, antibiotics, systematic hormones, douching process, contraceptives, frequent sexual intercourse, poor socio-economic status, and stress level can hamper typical vaginal ecosystem after a specific period. Sometimes, lactobacillus dominant vaginal microbiota is replaced by several harmful anaerobic and aerobic microbes (Sirota *et al*., 2014; Lykke *et al*., 2021). Babu *et al*. (2017) conducted one cross-sectional study in India by selecting 200 females (84 healthy + 116 infertile) aged 18 to 45 years. Study collected swab samples for microbiological analysis. This study showed a significant amount of *Lactobacillus* present in the vaginal samples among 27.8% (n= 40) of healthy women. Remaining 15.3% (n= 22), 11.1% (n= 16), and 8.3% (n= 12) of healthy women's vaginal samples were dominated by *Micrococcus*, *Enterococcus*, and *Staphylococcus*, respectively. At the same time, among the infertile patient group, 26.5% (n= 30), 23% (n= 26), and 14.1% (n= 16) of infertile women showed *Candida*, *Enterococcus*, and *Escherichia coli* dominated vaginal microbiota. A low level of *Lactobacillus* count was detected among infertile female patients. The result of the experiment also identified asymptomatic vaginosis among 27.6% of infertile patients and 7.1% of healthy females. This study recommended routine screening of the vaginal microbial system during infertility treatment (Babu *et al*., 2017). Some internal factors including age, immune power, hormonal status, and external factors such as infectious microbial exposure, antibiotic exposure, can facilitate the vaginal microbial disbalance process (dysbiosis). Vaginal microbiota dysbiosis is strongly associated with bacterial vaginosis, which further directly associated with women reproductive health disorders, HIV (human immunodeficiency virus), human papillomavirus (HPV), pelvic inflammatory disease risk. Some common

factors like douching, variation in intercourse, stress, race, regional disparity can also chance the vaginal microbiome composition (Babu *et al*., 2017). This review aims to summarize the adverse effect of different bacterial infections on female reproductive health and infertility.

CHLAMYDIA TRACHOMATIS

Chlamydia trachomatis is an obligate intracellular Gram-negative bacterium (Witkin *et al*., 2017). *C. trachomatis* (under the genus *Chlamydophila*) is responsible for Chlamydia, one kind of sexually transmitted infectious state. *C. trachomatis*, mainly serovars A-C are responsible for human blindness, serovars L1-L3 are responsible for lymphatic system infection and sexually transmitted infections *C. trachomatis* serovars D-K are especially responsible (Witkin *et al*., 2017). *C. trachomatis* can transmit directly through the vagina, oral and anal sex. The fetus can also get this infection during childbirth from the mother. This infection is widely prevalent around the United States as well as around the world. *C. trachomatis* is responsible for cervicitis, proctitis, urethritis, and trachoma, one kind of ocular infection that consequently leads to permanent blindness if remains untreated (CDCP, 2021b). Sexually transmitted infections like *C. trachomatis*, *Neisseria gonorrhoeae*, *Mycoplasma*, and *Treponema pallidum* can impair normal reproductive functions (Witkin *et al*., 2017). Persistent *C. trachomatis* infection can induce infertility and ectopic pregnancy chance. Young women usually get *Chlamydia* infection rapidly, increasing the chance of tubal infertility, pelvic inflammatory diseases, obstetrics consequences, and chronic pelvic pain. 10% to 15% of women infected with *Chlamydia* show symptomatic pelvic inflammatory disease (CDCP, 2021b; Srivastava *et al*., 2008). Most of the patients with chlamydial infection are asymptomatic. According to the Centers for Disease Control and Prevention, 4 million people were infected by *C. trachomatis* (CDCP, 2021b). In the United States, *Chlamydia* infection is the most common, followed by *Gonorrhea* infection cases. Centres for Disease Control and Prevention estimated 1808703 *Chlamydia* infection cases and 616392 *Gonorrhea* infection cases from fifty states of the United States and the District of Columbia in 2019 (CDCP, 2021c). Chlamydia infection also has some disadvantages, such as antibiotic resistance and side effects on urogenital microbiota (Mohseni *et al*., 2022; Debonnet *et al*., 2021). Parpillewar and Singh (2021) conducted one cross-sectional study to determine the prevalence of female infertility due to *C. trachomatis* infection. Seventy-five infertile women (with and without pelvic inflammatory disease) and 75 women with no infertility symptoms were enrolled in this study.

The study collected cervical swab samples from recruited women to detect *C. trachomatis* infection. A sudy detected 14 *C. trachomatis* infections from the infertile women group (42.85 were asymptomatic and 57.14 were symptomatic) and 4 from the control group. This study also agreed that *C. trachomatis* infection harms female fertility processes (Parpillewar and Singh, 2021). Sharaf *et al*. (2021) selected 50 female patients with primary and secondary infertility and 25 healthy pregnant ladies for their prospective randomized clinical study. The study analyzed the anti-chlamydial IgG level of the enrolled females and reported a higher level of IgG in infertile women (46%) compared to the normal pregnant women group (12%). Studies strongly suspect *C. trachomatis* is one of the major reasons for tubal factor infertility (Sharaf *et al*., 2021). A retrospective cohort study investigated 253 tubal factor infertile women (who went for tubal flushing) and showed that *C. trachomatis* infection significantly decreased the pregnancy chance and live birth rate. Therefore, the findings of the study suggest that *C. trachomatis* infection screening before the tubal flushing procedure (Kayiira *et al*., 2019). Another experimental study by Rashidi *et al*. (2013) was performed to measure the burden of *C. trachomatis* infection among fertile and non-fertile women group using PCR and ELISA techniques.

The study took 223 pregnant and 234 infertile women for the study purposes. The study showed that 12.4 % of the infertile women and 8.5% of the fertile women were *C. trachomatis* infected. Studies failed to show any significant difference in *C. trachomatis* infection rate between the pregnant and non-fertile groups of women. A study suggested the *C. trachomatis* infection diagnosis during infertility condition and recommended molecular techniques for determination of the infection (Rashidi *et al*., 2013). One more cross-sectional study in India investigated the *C. trachomatis* infection rate in infertile women. Mania-Pramanik *et al*. (2012) took 896 female patients and performed a PCR technique to determine the presence of *C. trachomatis* infection. Studies showed a significant negative effect of *C. trachomatis* infection on female fertility. The study also revealed that the *C. trachomatis* infection rate is much higher among women with ectopic pregnancy (25%) and infertility (18.6%). De Lima Freitas *et al*. (2011) used the polymerase chain reaction technique for *C. trachomatis* infection identification among 106 infertile women. After analysis study revealed that 52.8% of infertile women had *C. trachomatis* infection and out of this 51.8% were above 30 years old.

The study also reported that among 56 *C. trachomatis* infection cases, 55.4% were infertile permanently, and

16 % of women faced fetal death during pregnancy (de Lima Freitas *et al*., 2011). Another prospective study in India by Malik *et al*. (2009) also confirmed the presence of *Chlamydia* infection among women with secondary infertility. The study selected 40 women with secondary infertility and 30 healthy women as controls. Study results indicated that the *Chlamydia* infection rate among secondary infertile women was significantly high. The study also recommended the immunoglobulin G antibody detection process as a diagnosis tool for *Chlamydia* infection. The study also indicated the positive advantages of the ELISA technique such as inexpensiveness, ease of measurement, and early process for antibody and antigen diagnosis during Chlamydial infection (Siemer *et al*., 2008). Srivastava *et al*. (2008) collected vaginal swab samples from 133 infertile female patients to detect sexually transmitted infections. After analysis, the study identified 25 patients and 23 patients who had *C. trachomatis* and *Mycoplasma* infection, respectively. Apart from this, the study did not find any *Neisseria gonorrhoeae* and *Treponema pallidum* infection cases. At the end of the experiment, the study concluded that frequently, *C. trachomatis* and *Mycoplasma* infection detection is necessary in case of infertility treatment. India-based study with 368 female patients identified the effect of chlamydial heat shock proteins (cHSP) 60 and 10 on infertility and ectopic pregnancy. Chlamydial heat shock proteins (cHSP) 60 and 10 could stimulate the production of IL-10, Interferon-gamma, and tumor necrosis factor-alpha from cervical mononuclear cells, and this phenomenon is related to *C. trachomatis* infection-mediated infertility and ectopic pregnancy.One retrospective study by Siemer *et al*. (2008) (191 with primary and secondary infertility, 248 healthy pregnant women) women did not show any significant association between *C. trachomatis* infection and infertility rate, but this study observed infection-specific IgG and IgA antibodies within both primary and secondary infertile women. Al-Ramahi *et al*. (2008) conducted one Jordan-based prospective controlled study. The study aimed to check the burden of *C. trachomatis* infection among infertile women over a specific area. The study recruited 146 healthy women as control and 152 infertile patients and collected endocervical swab samples for infection detection by polymerase chain reaction technique. No significant effect of *C. trachomatis* infection on infertility rate was reported. Malik et al. (2009) conducted one study to observe the effect of *C. trachomatis* on female infertility. The study recruited 110 women with primary and secondary infertility and 30 healthy pregnant women as control. The findings of the study stated that 28.1% of the infertile women and 3.3% of the control population had *Chlamydia* infection (detection of *C. trachomatis* infection). The study detected more *C. trachomatis* infection cases from the infertile women group, especially from asymptomatic cases.

The study recommended an early screening process for *C. trachomatis* infection to establish preventive, therapeutic measures against this infection as soon as possible (Malik *et al*., 2009). Lucisano *et al*. (1992) isolated *C. trachomatis* from 105 women who underwent laparoscopy. The study collected urethral, cervical, endometrial, and peritoneal samples for *C. trachomatis* isolation. Out of 42 tubal infertilities, 41 were unexplained infertility, 4 were salpingitis, and 18 were endometriosis cases 13, 5, 1, and 1 female patient were infected with *C. trachomatis* infection, respectively. However, the study confirmed the association of *C. trachomatis* infection with tubal damage (Lucisano *et al*., 1992).

MYCOPLASMA GENITALIUM

Mycoplasma comes under the Mollicutes class (Ljubin-Sternak and Meštrović, 2014). Genus *Mycoplasma* consists of more than 100 different strains (Mycoplasma infections-statpearls- NCBI Bookshelf, 2022). *Mycoplasma genitalium* infection can lead to abnormal inflammatory conditions like cervicitis, urethritis, salpingitis, pelvic inflammatory disease (Afrasiabi et al., 2015), and endometritis and ultimately increase the chances of getting female infertility. *M. hominis* infection is associated with bacterial vaginosis (Plummer *et al*., 2021), pyelonephritis, cervicitis, tubal inflammation, pelvic inflammatory disease, endometritis, and postpartum septicemia. Another genital mycoplasma, *Ureaplasma urealyticum* infection can induce preterm delivery, bacterial vaginosis, cervicitis, urethritis, and chorioamnionitis (Afrasiabi et al., 2015). Women's reproductive health can be altered due to various genital mycoplasma infections. Some species like *M. genitalium*, *Mycoplasma hominis*, *Ureaplasma parvum*, *M. spermatophilum*, *M. primatum*, *M. penetrans*, *Ureaplasma urealyticum* are commonly known as genital *Mycoplasma*.

Ma *et al*. (2021) studied the potential role of genital mycoplasmas on female infertility and pregnancy outcomes. Based on the previous 35 electronic databases, the study concluded that genital *Mycoplasma*, *M. genitalium* infection was one of the significant causative factors for preterm delivery and female infertility. However, there was no role of *M. genitalium* infection in spontaneous abortion. Whereas, *M. hominis* increased female infertility, stillbirth, and preterm membrane rupture chances. The study found no potential role of *Ureaplasma urealyticum* on female infertility (Ma *et al*., 2021). Peipert *et al*. (2021) used proportional hazards models and found that age, low socio-economic status, black race,

and previous record of *M. genitalium* infection could lower the chances of conception. The study confirmed the impact of *M. genitalium* infection on fertility and conception rate (delayed conception) by performing a serological analysis process on 461 participants (Peipert *et al*., 2021). Tantengco *et al*., (2021) performed a meta-analysis to confirm the impact of genital *Mycoplasma* on female infertility status. This meta-analysis also selected *M. genitalium*, *M. hominis*, *Ureaplasma parvum*, and *Ureaplasma urealyticum* as potent genital mycoplasmas. This meta-analysis confirmed the potential effect of *M. genitalium*, *M. hominis*, and *Ureaplasma urealyticum* infection on female reproductive health (female infertility) and suggested *Mycoplasma* diagnosis during infertility management (Tantengco *et al*., 2021). Doroftei *et al*. (2021) proved the negative impact of *M. genitalium* infection on women's fertility process. This study recruited 51 infertile patients and 23 females with normal fertility. 19.6 % and 4.4% of infertile and fertile women had *M. genitalium* infection in their cervical canal, respectively. The study also identified *M. genitalium* infection in the abdominal cavity of 5.8% of infertile women (Doroftei *et al*., 2021). Another related study also checked the burden of *Ureaplasm aurealyticum* and *M. hominis* infection within 411 registered infertile women. As per the study, 28.46%, 2.91%, and 0.48% of the infertile women had *Ureaplasm aurealyticum*, coinfection, and *M. hominis* infection respectively (Doroftei *et al*., 2021).

One Iran-based descriptive study investigated the infection rate of *C. trachomatis*, *M. genitalium*, and *Neisseria gonorrhoea* within infertile women. The study collected 65 infertile women to collect vaginal swab samples from them. PCR and DNA extraction techniques were performed and it ultimately found that out of a total of 65 female samples, 23 patients were infected with bacterial infection (2 patients had mixed infection). 16.9%, 13.8%, and 6.2% of infertile patients had *M. genitalium*, *C. trachomatis*, and *Neisseria gonorrhea* infection, respectively. The study confirmed the significant presence of *C. trachomatis*, *M. genitalium*, *Neisseria gonorrhea* infections among women with infertility (Sameni *et al*., 2022). Another experimental study was conducted to determine *M. genitalium*, *M. hominis*, and *Ureaplasma urealyticum* infection prevalence among infertile women candidates. The study recruited 104 infertile women and collected their cervical swab samples for further analysis. Multiplex-PCR techniques detected *Ureaplasma urealyticum* infection among 37.5% of the selected females. Only 2.9% had *M. genitalium*, *M. hominis* infection. The study found no association between infertility and the patient's education, age, employment nature, first intercourse age, or abortion history (Mousavi, *et al*., 2014).

Baczynska *et al*. (2006) conducted an *in vitro* experiment. They detected the moderate effect of *M. genitalium* infection on normal human fallopian tubes. However, they observed massive alteration of the epithelium of normal human fallopian tubes after exposure to *C. trachomatis* and *Neisseria gonorrhea* infection (Baczynska *et al*., 2006). Baczynska *et al*. (2005) suspected *M. hominis* infection as one of the major risk factors for infertility due to damaged fallopian tubes. Their study analyzed sera for *M. hominis* specific antibody detection by selecting 304 infertile women. This study identified *M. hominis*-specific antibodies in the sera sample of 97 infertile women. The result of the study indicated a potent relation between insufficient fallopian tube passage-related infertility and *M. hominis* infection rate (Baczynska *et al*., 2005). Figure 1 lists seven significant *Mycoplasma* strains together with each one's harmful characteristics.

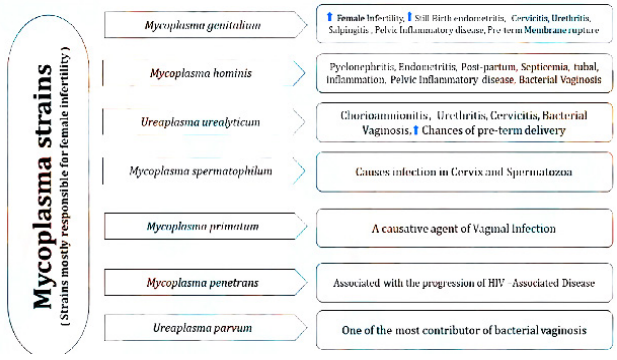

Fig. 1. Different strains of *Mycoplasma* and their pathological role.

BACTERIAL VAGINOSIS

A change in the vaginal microbiota's delicate equilibrium is the main contributor of BV. The vagina naturally harbours many microorganisms, including helpful lactobacilli that support the vagina's acidic pH balance and inhibit the growth of potentially hazardous bacteria. Although BV is not considered an STI, it is more prevalent in people who engage in sexual activity, particularly those with several sexual partners. Frequent douching increases the risk of BV and disturbs the vaginal flora. Soaps, scented goods, or vaginal deodorants can interfere with the typical vaginal environment. Smoking has been linked to a higher chance of developing BV. According to certain studies, using an intrauterine device (IUD) is associated with BV.

Typical signs included in vaginal discharge that is greyish-white, a strong, fishy smell, especially after sexual activity, itching or discomfort in the vagina and a

burning feeling when urinating. A combination of clinical evaluation and laboratory tests are used to diagnose BV. Medical history and clinical exam, evaluation of vaginal discharge, and microscopic analysis of the vaginal discharge are some common methods to detect BV. Treatment including antimicrobial therapy, which tries to reduce the excess of dangerous bacteria and reestablish the normal vaginal flora is commonly use. During bacterial vaginosis, lactobacillus-dominated vaginal microbiota is replaced by a wide range of harmful pathogens like *Gardenella vaginalis*, *Mobiluncus*, *Atopobium*, *Prevotella*, *Dialister*, *Mycoplasma*, *Streptococcus*, *Ureaplasma*, *Bacteroides* and many more. These anaerobes decrease antimicrobial peptides amount and normal vaginal pH by inhibiting lactic acid concentration and exhibit the level of short-chain fatty acids (butyrate, acetate, succinate, propionate) and immune mediators (interferon, IL- 2, IL- 6, IL- 8, IL- 10, IL-1β, TNFα RANTES) (Amabebe and Anumba, 2018; Sirota *et al*., 2014). Vaginal amylase can split complex carbohydrates into glycogen, serving as bacterial food for normal vaginal bacterial growth and survival. The vaginal amylase content gets low during bacterial vaginosis; therefore, normal bacteria cannot grow and survive. Additionally, the human body becomes deprived of antimicrobial peptides during bacterial vaginosis. Bacterial vaginosis can trigger female infertility by elevating inflammatory responses, hampering immune system potentiality, damaging normal vaginal cells and sperm, damaging the production of cervical mucus at the time of ovulation, and clogging the fallopian tube by infections mediated scar tissues, decreasing sperm and egg meeting process in a fallopian tube (Nunn *et al*., 2020; APA, 2022). About 67 species are available that cause bacterial vaginosis. Name of some common bacterial vaginosis-causing bacteria are *Gardnerella vaginalis, Dialister* spp., *Megasphaera* spp., *Atopobiumvaginae*, *Sneathiaamnii*, *Sneathiasanguinegens*, *Porphyromonas* spp., *Prevotella* spp., *Mobiluncus* spp. (Ravel *et al*., 2021). Many studies proved the association between idiopathic infertility and bacterial vaginosis, elevated vaginal pro-inflammatory cytokines (IL- 8, IL- 1 β) levels.

Bacterial vaginosis is directly associated with poor reproductive health, tubal factor infertility, implantation failure, pregnancy loss, and other sexually transmitted infection's chance. At the same time, some infectious conditions in the female reproductive organs, like pelvic inflammatory disease, Bacterial vaginosis, (Mania-Pramanik *et al*., 2012), and endometritis, can put a woman into various reproductive health-related issues, including infertility. A common symptom of bacterial vaginosis is whitish or grey vaginal discharge with a fishy odour. Other symptoms are dyspareunia, dysuria, vaginal pruritus, etc. Patients with bacterial vaginosis are more prone to get other sexually transmitted infections like gonorrhea, and chlamydia; even pregnant women with bacterial vaginosis may have a higher risk of preterm delivery. Bacterial vaginosis is not transmitted from person to person. Bacterial vaginosis induces endotoxin secretion, further triggering prostaglandin and cytokine production in the vagina. Simultaneously, bacterial vaginosis inhibits the potentiality of the host leukocytes against infectious diseases. The presence of clue cells, the cervix's epithelial cells is the main indicator of bacterial vaginosis. Douching, antibiotic treatment, multiple sexual partners, frequent use of intrauterine devices, and cigarette smoking contribute to bacterial vaginosis (Kairys and Garg, 2022). Wee *et al*. (2018) performed one initial pilot study (case-control study) to check the vaginal and cervical microbiota composition of women with infertility and normal fertile women. The study selected 31 women (15 infertile and 16 fertile) for sample collection and performed 16 rRNA gene amplicon sequencing techniques to analyze the collected samples. The study stated that *Ureaplasma* and *Gardenella* were most abundantly present in the vaginal and cervical samples of the selected infertile women, respectively. Babu *et al*. (2017) checked the difference between the prevalence of bacterial vaginosis in healthy and infertile women by establishing one cross-sectional study in India. The study considered only 200 women (84 healthy females, 116 infertile females) in the reproductive age group for the study purpose. Studies showed a greater number of *Lactobacillus* (27.8%) within the vaginal flora of the healthy women group. At the same time, the vaginal flora of the infertile women group was dominated by *Candida* spp. (26.5%), *Enterococcus* (23%), *Escherichia coli* (14.1%). The study detected the significant presence of bacterial vaginosis and asymptomatic vaginosis among the infertile women population compared to healthy females. Systematic literature reviews from PubMed, CINAHL, EMBASE, ISI Web of Knowledge, and Cochrane Library showed that women with bacterial vaginosis are more prone to get tubal factor infertility. The incidence rate of bacterial vaginosis was higher among tubal factor infertile women and was often associated with preclinical pregnancy loss (van Oostrum *et al*., 2013). A cohort study once conducted by Salah *et al*. (2013) and the study considered 382 asymptomatic fertile women (control) and 874 infertile women to investigate the presence of bacterial vaginosis in the vaginal samples of the mentioned women and the effect of bacterial vaginosis on fertility and pregnancy rate. The study's findings showed that 45.5% of the infertile women had bacterial vaginosis, whereas only 15.4% of the fertile women had bacterial vaginosis. As per the regression model, bacterial vaginosis is one of the major significant

factors for adverse pregnancy outcomes and faulty fertility processes (mainly unexplained infertility). Casari *et al*. (2010) investigated the present rate of pathogens among asymptomatic infertile and symptomatic fertile women (a total of 952 patients). The study investigated the genital discharge of recruited females and showed that 19.7%, 12.1%, and 8.6% of infertile patients had *Gardenella vaginalis*, *Enterococci*, and *Streptococcus agalactiae* in their genital discharge, respectively. A study mentioned the decreased number of vaginal *Lactobacillus* and increased polymorphonucleates number as indicative parameters to determine female urogenital tract health status. Nwaziri *et al*. (2010) conducted one *in vivo* study to detect the *Gardenella vaginalis* effects on pregnancy and infertility by using albino rat models. A study found a 20-40% decrease in impregnation and a 70-80% reduction of offspring production ability by rats after being infected with *Gardenella vaginalis* (105CFU/ml). This study concluded that *Gardenella vaginalis* negatively affects rats normal fertility process and pregnancy outcomes. One experimental study also investigated the effect of vaginal microbiota on early pregnancy failure and conception rate in women who went for in-vitro fertilization (IVF). The study only enrolled 91 female patients for this study. The study analyzed and confirmed that IVF patients with bacterial vaginosis and a low amount of hydrogen peroxide synthesizing *Lactobacillus* within vaginal microbiota are more vulnerable to early pregnancy loss and lower conception chance (Eckert *et al*., 2003). One cross-sectional study with 749 confirmed bacterial vaginosis was more common among tubal infertile women than endometriosis, male factors infertility, and unexplained infertility (Wilson *et al*., 2002). Spandorfer *et al*. (2001) conducted one masked study with 331 IVF patients, and the findings showed that bacterial vaginosis was strongly linked with a higher level of IL-1 beta and IL-8 in the cervix. The pro-inflammatory cytokines production by the vaginal microbial community might increase the risk of idiopathic infertility.

CHRONIC ENDOMETRITIS

Chronic endometritis (CE) is an inflammatory condition where plasma cells are detectable inside the endometrial stroma during diagnosis. Alteration of the endometrial microbiota composition leads to chronic endometritis (CE), which enhances the chance of bacterial vaginosis and preterm delivery. Intracellular gram-negative microbes like *Mycoplasma*, *Enterococcus faecalis*, *Chlamydia*, *Ureaplasma*, *Staphylococcus*, and *Escherichia coli* are primarily responsible for altering endometrial microbiota. Low *Lactobacillus* content is very natural within the endometrial cavity during chronic endometritis. Buzzaccarini *et al*. (2020) summarized fifteen original electronic articles stating that CE can consequently inhibit fertility rate in females by infection-mediated inflammation, altering the endometrium microbial environment, altering the permeability of the endometrium vascular system, altering leukocytes and cytokines expression, and injuring viability of the embryo. Symptomatic and asymptomatic chronic endometritis is directly related to pregnancy failure, miscarriage, and other associated gynaecological problems. Chronic endometritis has been found in approximately 40% of infertile patients (Holzer *et al*., 2021). Elnashar (2021) suggested chronic endometritis diagnosis in infertile females to improve the clinical management of asymptomatic patients in whom chronic endometritis is not suspected or diagnosed. Many immunocompetent cells in the endometrium can regulate immune and inflammatory associated responses and trophoblast implantation. However, immunogenic cell alteration is common during endometritis, further responsible for immune cell infiltration, hamper immune and inflammatory responses of endometrial cells, and ultimately, embryo implantation and growth endometrium (Ravel *et al*., 2021). For their study, Hirata *et al*. analyzed 53 female patients (26 were diagnosed with CE, and 27 patients had no CE during diagnosis).

Studies showed that patients with CE were more prone to miscarriage risk. CE also harmed the live birth rate and pregnancy rate among selected women. Live birth rate pregnancy and miscarriage rate among female patients with CE were 7.7%, 30.8%, and 75%, respectively, whereas live birth rate pregnancy and miscarriage rate among female patients without CE were 51.9%, 63.0%, and 17.7%, respectively. This study advised specific diagnostic criteria for CE: the detection of > = one plasma cell in 10 power fields (Hirata *et al*., 2021). Wiesenfeld *et al*. (2012) conducted one prospective observational cohort study with 418 women. Those selected women were either at risk of gonorrhea, Chlamydia, or bacterial vaginosis. No women had pelvic inflammatory disease. A study showed a 40% reduction in the pregnancy rate among women with subclinical pelvic inflammatory disease (endometritis) compared to women without subclinical pelvic inflammatory disease. The study report showed that *Chlamydia* and *Neisseria gonorrhoeae* infection without the subclinical pelvic inflammatory disease did not affect infertility risk. The study also suggested that recent preventive therapies for sexually transmitted infections related to infertility problems are insufficient to prevent infertility rate (Wiesenfeld *et al*., 2012). Li *et al*. (2017) selected 100 infertile women for a prospective, monocentral pilot study. The expert group

detected chronic endometritis among 13 infertile women (13%), and they had associated unilateral or bilateral fallopian tube blockage and endometriosis. The study specified that stages of endometriosis depended upon the presence of syndecan-1- 1 (CD138) positive plasma cells within endometrial tissue. One randomized controlled trial showed a 2.8% rate of chronic endometritis among 606 asymptomatic infertile women using a biopsy test. The study stated that chronic endometritis had minimal effect on the reproductive outcome of infertile women after IVF/ICSI treatment (Kasius *et al*., 2011).

ENDOMETRIOSIS

Endometriosis is one type of vaginal microbiota dysbiosis condition among reproductive-aged women, further contributing to women's infertility. 40% to 50% of the reproductive age female with endometriosis may face infertility in their reproductive life. In endometriosis, endometrial tissue formation is common outside of the uterine cavity. Endometriosis is one of the major causes of infertility (Kasius *et al*., 2011). Nearly 30 to 71% of infertile women are generally detected with endometriosis, and approximately 30 to 50% of the women with endometriosis are diagnosed as infertile (López-Moreno and Aguilera, 2021). Microbes responsible for endometriosis are *Gardnerella*, *Prevotella*, *Staphylococcus*, *Escherichia coli*, *Cornybacterium*, *Actinomyces*, *Enterococcus*, *Streptococccus*, *Propionibacterium* (Tomaiuolo *et al*., 2020). Shi *et al*. (2021) took 226 female infertile patients (176 patients completed the experiment) and found that both adenomyosis and endometriosis harmed reproductive activity and pregnancy outcome. This study also suggested the IVF technique and laparoscopic surgery to improve the pregnancy outcome in patients with adenomyosis and endometriosis. Corachán *et al*. (2021) identified some significant consequences within female patients with endometriosis such as dysregulation of the cell cycle (impaired quality of oocyte and embryo, poor folliculogenesis process and IVF outcomes), oxidative stress and inflammatory response, biosynthesis and metabolism of steroid (impaired quality of oocyte and embryo), and angiogenesis process into the oocyte and follicular environment by using 123 endometriosis-related previous data (from 1992- 2020) available in PubMed database. This study indicated endometriosis as one of the main reasons for female infertility problems due to those, as mentioned above, endometriosis-associated clinical factors. Campisciano *et al*. (2017) took 27 infertile female patients and 69 fertile average women and collected cervical-vaginal fluid samples from them for the experiment. A study found that endometriosis is the primary cause of infertility among selected infertile women group, and they did not have normal vaginal Lactobacillus in their vaginal microbiota. After performing the V3-16rDNA sequencing technique study, they indicated Lactobacillus as a biomarker for a normal healthy vaginal ecosystem.

PELVIC INFLAMMATORY DISEASE

In pelvic inflammatory disease, the inflammatory condition has been seen in the uterus, fallopian tubes and ovaries, and pelvic part of the female reproductive system due to pathogenic infection. This inflammation-causing infection can enter through the vagina and infect up to the endometrium portion or beyond. Sometimes, clinical symptoms of pelvic inflammatory disease are asymptomatic. However, this inflammatory condition usually leads to several clinical consequences like parametritis, endometritis, oophoritis, salpingitis, pelvic peritonitis, tubo-ovarian abscess, perihepatitis, and ovarian cancer. Pelvic inflammatory disease is becoming a public health burden because nearly 30% of female infertility cases and 50% of ectopic pregnancy cases are due to pelvic inflammatory disease (Al-Kuran *et al*., 2021). Pelvic inflammatory disease (PID) is a serious infectious disease that directly affects women's fertility capacity (Witkin *et al*., 2017). Pelvic inflammatory disease (PID) is an infection (mainly sexually transmitted infections like *C. trachomatis* and *Neisseria gonorrhoeae*) mediated inflammatory condition among women that starts to develop from the lower genital tract and gradually spread to the uterus, fallopian tube, and ovaries (DM, 2022). Cervicitis is also an inflammatory condition that affects uterine endocervix epithelium. It can happen from both infectious and non-infectious sources (Iqbal *et al*., 2025). Infection in the female reproductive organs (endometrium, fallopian tube, peritoneum) leads to pelvic inflammatory disease. This infection state often occurs by sexually transmitted infectious pathogens invading Gonorrhea, Chlamydia. Pathogens transmit to the upper genital tract through the vagina and cervix.

According to the National Health and Nutrition Examination Survey, 2013-2014 report, the lifetime pelvic inflammatory disease rate among sexually active women in the United States is 4.4%, almost 2.5 million females across the United States. This number is far higher than the 2001-2004 report (21.2 million women). The pelvic inflammatory disease ultimately leads to female infertility, lower pregnancy outcomes, pelvic pain, ectopic pregnancy, and many other reproductive health-related issues (Kreisel *et al*., 2017; CDCP, 2020). Anti-cancerous role of Flavonoids, Catechin, β-sitosterol, and Lignin Glycosides from *Saracaasoca* (Ashoka) have been discussed with

reference to the female reproductive system (Ahmad and Ghosh, 2022). A group of researchers from different countries had worked on the Antineoplastic action of sulforaphane on HeLa cells by modulation of signaling pathways and epigenetic pathways. It showed how cancer call can be controlled by chemical interventions (Sundaram *et al*., 2021). Nanotechnology can completely alter cancer treatment by providing cutting-edge medication delivery, diagnostics, and imaging options, among other problems. While nanotechnology cannot be used to cure cancer directly through food, it can be used to improve the efficacy of cancer treatment when added to medicinal formulations or medical equipment (Ahmad and Ghosh, 2020).

CONCLUSION

Microbial dysbiosis may stimulate various kinds of bodily malfunctions, including hormonal dysfunction, metabolic dysfunction, reproductive organ dysfunction, and so on. From this review study on the female infertile condition, vaginal microbiota significantly contributes. The vaginal microbiota is essential in maintaining female reproductive health and preventing gynaecological problems. Generally, a normal healthy vaginal contains a very few pathogenic microbes. Lactobacillus has a direct role behind this because *Lactobacillus* maintains vaginal pH around 3.5 to 4.5 by producing lactic acid which helps to reduce pathogens (*Gardnerella vaginalis*, *Pepto-streptococci*, *Anaerobic rods*, and *mycoplasma* species) growth and multiplication. Antibacterial compounds like H_2O_2 bacteriocins and bacteriostatic compounds are also produced by *Lactobacillus*, which helps in the bacterial lysis process. *Lactobacillus* can prevent various pathogenic infection-related gynaecological problems such as pelvic inflammatory disease, endometriosis, chronic endometritis, and cancer. Vaginal microbial dysbiosis and exposure to any pathogenic microorganisms can lead to infertility. This review paper concluded that pathogens, like sexually transmitted infection-causing bacteria (*Treponema pallidum*, *Neisseria gonorrhoeae*, *Mycoplasma*, *C. trachomatis*) and a wide range of reproductive organs associated with inflammatory conditions like pelvic inflammatory disease, endometriosis, and chronic endometritis, may hamper normal fertility process in the women population. The location of the onset or occurrence of bacterial vaginosis is depicted in Figure 2. Figure 3 demonstrates how various microbial strain infections cause an imbalance that results in a condition known as microbial dysbiosis in the vagina.

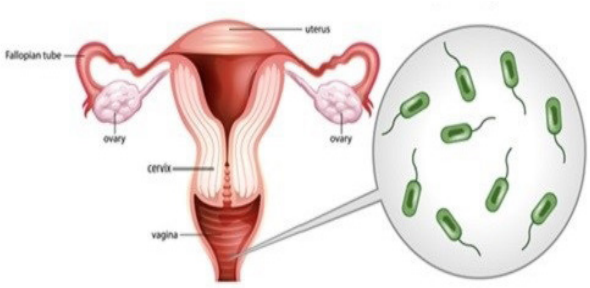

Fig. 2. Place of infestation of bacterial vaginosis.

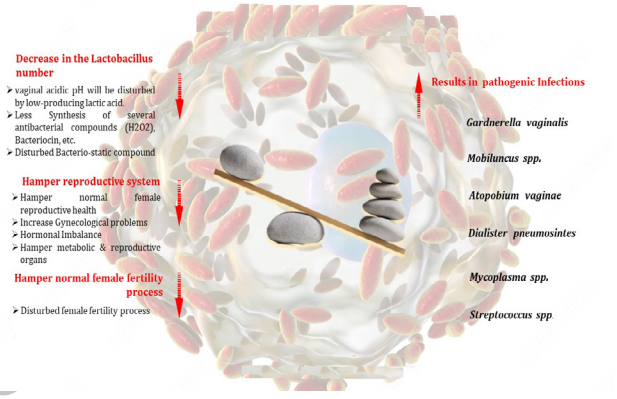

Fig. 3. Microbial dysbiosis in vagina.

DECLARATIONS

Acknowledgments

We, Ahmad, Husain, and Pritha, express our profound gratitude to the Almighty, whose blessings have inspired, strengthened, and sustained us throughout this endeavor. We hope that our work will contribute meaningfully to scientific understanding and the advancement of society. We also extend our sincere thanks to the Deanship of Research and Gr`aduate Studies at King Khalid University, KSA, for their financial support.

Funding

All authors, especially Dr. Izhar Husain, would like to extend their gratitude to the Deanship of Research and Graduate Studies at King Khalid University, KSA, for their financial support through project number RGP1/331/45.

Statement of conflict of interest

The authors have declared no conflict of interest.

REFERENCES

Afrasiabi, S., Moniri, R., Samimi, M., Khorshidi, A. and Mousavi, S.G., 2015. The prevalence of

endocervical *Chlamydia trachomatis* infection among young females in Kashan, Iran. *Jundishapur J. Microbiol.*, **8**: e15576. https://doi.org/10.5812/jjm.8(4)2015.15576

Ahmad, S.R. and Ghosh, P., 2022. A systematic investigation on flavonoids, catechin, β-sitosterol, and lignin glycosides from *Saracaasoca* (Ashoka) having anti-cancer and antioxidant properties with no side effects. *J. Indian Chem*., **99**: 1-10. https://doi.org/10.1016/j.jics.2021.100293

Ahmad, S.R. and Ghosh, P., 2020. Application of nanotechnology in the food industry and its potential toxicity-related health issues. *Indian J. Pure appl. Biosci.,* **8**: 678-689. https://doi.org/10.18782/2582-2845.8336

Al-Kuran, O., Al-Mehaisen, L., Alduraidi, H., Al-Husban, N., Attarakih, B., Sultan, A., Othman, Z., AlShárat, S., AlHilali, S., Alkouz, N., Alibrahim, N. and Al-Musallam, W., 2021. How prevalent are symptoms and risk factors of pelvic inflammatory disease in a sexually conservative population? *Reprod. Hlth.*, **18**. https://doi.org/10.1186/s12978-021-01155-2

Al-Ramahi, M., Mahafzah, A., Saleh, S., Fram, K., 2008. Prevalence of *Chlamydia trachomatis* infection in infertile women at a university hospital in Jordan. *East Mediterr. Hlth. J.,* **14**:1148-1154.

Amabebe, E. and Anumba, D.O., 2018. The vaginal microenvironment: The physiologic role of *Lactobacilli*. *Front. Med.*, **5**. https://doi.org/10.3389/fmed.2018.00181

APA, 2022. *Bacterial vaginosis (BV) and fertility*. American Pregnancy Association. https://americanpregnancy.org/getting-pregnant/bacterial-vaginosis-and-fertility/

Babu, G., Singaravelu, B.G., Srikumar, R., Reddy, S.V. and Kokan, A., 2017. Comparative study on the vaginal flora and incidence of asymptomatic vaginosis among healthy women and in women with infertility problems of reproductive age. *J. Clin. Diagn. Res.,* **11**: DC18-DC22. https://doi.org/10.7860/JCDR/2017/28296.10417

Baczynska, A., Friis, S.H., Fedder, J., Birkelund, S. and Christiansen, G., 2005. The use of enzyme-linked immunosorbent assay for detection of *Mycoplasma hominis* antibodies in infertile women serum samples. *Hum. Reprod.*, **20**: 1277–1285. https://doi.org/10.1093/humrep/deh780

Baczynska, A., Funch, P., Fedder, J., Knudsen, H.J., Birkelund, S. and Christiansen, G., 2006. Morphology of human fallopian tubes after infection with *Mycoplasma genitalium* and *Mycoplasma hominis in vitro* organ culture study. *Hum. Reprod.*, **22**: 968–979. https://doi.org/10.1093/humrep/del455

Buzzaccarini, G., Vitagliano, A., Andrisani, A., Santarsiero, C.M., Cicinelli, R., Nardelli, C., Ambrosini, G. and Cicinelli, E., 2020. Chronic endometritis and altered embryo implantation: A unified pathophysiological theory from a literature systematic review. *J. Assist. Reprod. Genet.*, **37**: 2897–2911. https://doi.org/10.1007/s10815-020-01955-8

Campisciano, G., Florian, F., D'Eustacchio, A., Stanković, D., Ricci, G., De Seta, F. and Comar, M., 2017. Subclinical alteration of the cervical-vaginal microbiome in women with idiopathic infertility. *J. Cell. Physiol.*, **232**: 1681–1688. https://doi.org/10.1002/jcp.25806

Casari, E., Ferrario, A., Morenghi, E. and Montanelli, A., 2010. *Gardnerella, Trichomonas vaginalis, Candida, Chlamydia trachomatis, Mycoplasma hominis* and *Ureaplasm aurealyticum* in the genital discharge of symptomatic fertile and asymptomatic infertile women. *New Microbiol.*, **33**: 69-76.

CDCP, 2020. *CDC- bacterial vaginosis statistics*. Centers for Disease Control and Prevention. https://www.cdc.gov/std/bv/stats.htm

CDCP, 2021a. *Infertility*. Centers for Disease Control and Prevention. https://www.cdc.gov/reproductivehealth/infertility/index.htm

CDCP, 2021b. *Detailed Std facts Chlamydia*. Centers for Disease Control and Prevention. https://www.cdc.gov/std/chlamydia/stdfact-chlamydia-detailed.htm

CDCP, 2021c. *Infertility and stds STD information from CDC*. Centers for Disease Control and Prevention. https://www.cdc.gov/std/infertility/default.htm

Corachán, A., Pellicer, N., Pellicer, A. and Ferrero, H., 2021. Novel therapeutic targets to improve IVF outcomes in endometriosis patients: A review and future prospects. *Hum. Reprod. Update*, **27**: 923–972. https://doi.org/10.1093/humupd/dmab014

De Lima Freitas, N.S., Borborema-Santos, C.M., Barroso Serrão das Neves, D., Costa de Oliveira, C.M., Dutra-Ferreira, J.R. and Astolfi-Filho, S., 2011. High prevalence detection of *Chlamydia trachomatis* by polymerase chain reaction in endocervical samples of infertile women attending University Hospital in Manaus-Amazonas, Brazil. *Gynecol. Obst. Invest.*, **72**: 220–226. https://doi.org/10.1159/000324798

Debonnet, C., Robin, G., Prasivoravong, J., Vuotto, F., Catteau-Jonard, S., Faure, K., Dessein, R.

and Robin, C., 2021. Infection à *Chlamydia trachomatis*: mise au point (Update of *Chlamydia trachomatis* of infection). *Gynécol. Obstét. Fertil. Sénol.*, **49**: 608-616. https://doi.org/10.1016/j.gofs.2021.01.003

DM, J.L., 2022. *Pelvic inflammatory disease*. National center for biotechnology information. https://pubmed.ncbi.nlm.nih.gov/29763134/

Doroftei, B., Ilie, O.D., Armeanu, T., Anton, E., Scripcariu, I. and Maftei, R., 2021. The prevalence of *Ureaplasmaurealyticum* and *Mycoplasma hominis* infections in infertile patients in the Northeast region of Romania. *Medicina*, **57**: 211. https://doi.org/10.3390/medicina57030211

Eckert, L.O., Moore, D.E., Patton, D.L., Agnew, K.J. and Eschenbach, D.A., 2003. Relationship of vaginal bacteria and inflammation with conception and early pregnancy loss following *in-vitro* fertilization. *Infect. Dis. Obst. Gynecol.*, **11**: 11–17. https://doi.org/10.1155/S1064744903000024

Elnashar, A.M., 2021. Impact of endometrial microbiome on fertility. *Middle East Fert. Soc. J.*, **26**. https://doi.org/10.1186/s43043-020-00050-3

Hirata, K., Kimura, F., Nakamura, A., Kitazawa, J., Morimune, A., Hanada, T., Takebayashi, A., Takashima, A., Amano, T., Tsuji, S., Kaku, S., Kushima, R. and Murakami, T., 2021. Histological diagnostic criterion for chronic endometritis based on the clinical outcome. *BMC Womens Hlth.*, **21**. https://doi.org/10.1186/s12905-021-01239-y

Holzer, I., Ott, J., Kurz, C., Hofstetter, G., Hager, M., Kuessel, L. and Parry, J.P., 2021. Is chronic endometritis associated with tubal infertility? A prospective cohort study. *J. Minimally Invasive Gynecol.*, **28**: 1876–1881. https://doi.org/10.1016/j.jmig.2021.04.011

Hossein, R.B., Shams, S., Shariat, M., Kazemi, J.H., Mohebi, M. and Haghollahi, F., 2017. Evaluation of serum hepcidin and iron levels in patients with PCOS: A case-control study. *J. Endocrinol. Invest.*, **40**: 779–784. https://doi.org/10.1007/s40618-017-0632-z

Iqbal, U., Carlson, K. and Wills, C., 2025. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 Jan. Available from: https://www.ncbi.nlm.nih.gov/books/NBK562193/

Kairys, N. and Garg, M., 2022. Bacterial vaginosis statpearls NCBI bookshelf. https://www.ncbi.nlm.nih.gov/books/NBK459216/

Kalidasan, M., Poulpunitha, S. and Veeramani, P., 2020. Infertility: An alarming situation in India. *Int. J. Sci. Technol. Res.*, **9**: 2606-2609.

Kasius, J.C., Fatemi, H.M., Bourgain, C., Sie-Go, D.M.D.S., Eijkemans, R.J.C., Fauser, B.C., Devroey, P. and Broekmans, F.J.M., 2011. The impact of chronic endometritis on reproductive outcome. *Fert. Sterility*, **96**: 1451–1456. https://doi.org/10.1016/j.fertnstert.2011.09.039

Kayiira, A., Zaake, D., Lwetabe, M.W. and Sekweyama, P., 2019. Impact of genital *Chlamydia trachomatis* infection on reproductive outcomes among infertile women undergoing tubal flushing: A retrospective cohort at a fertility centre in Uganda. *Fert. Res. Pract.*, **5**: 16(2019). https://doi.org/10.1186/s40738-019-0069-5

Kreisel, K., Torrone, E., Bernstein, K., Hong, J. And Gorwitz, R., 2017. Prevalence of pelvic inflammatory disease in sexually experienced women of reproductive age United States, 2013–2014. *Morbidity Mortal. Weekly Rep.*, **66**: 80–83. https://doi.org/10.15585/mmwr.mm6603a3

Li, M., Zhang, X., Huang, K., Qiu, H., Zhang, J., Kang, Y. and Wang, C., 2017. Presence of *Chlamydia trachomatis* and *Mycoplasma* spp., but not *Neisseria gonorrhoeae* and *Treponema pallidum*, in women undergoing an infertility evaluation: High prevalence of tetracycline resistance gene Tet(m). *AMB Express*, **7**. https://doi.org/10.1186/s13568-017-0510-2

Ljubin-Sternak, S. and Meštrović, T., 2014. *Chlamydia trachomatis* and genital Mycoplasmas: Pathogens with an impact on human reproductive health. *j. Pathog.*, **2014**: 1–15. https://doi.org/10.1155/2014/183167

López-Moreno, A. and Aguilera, M., 2021. Vaginal probiotics for reproductive health and related dysbiosis: Systematic review and meta-analysis. *J. Clin. Med.*, **10**: 1461. https://doi.org/10.3390/jcm10071461

Lucisano, A., Morandotti, G., Marana, R., Leone, F., Branca, G., Dell'Acqua, S. and Sanna, A., 1992. CHLAMIDIAL genital infections and laparoscopic findings in infertile women. *Eur. J. Epidemiol.*, **8**: 645–649. https://doi.org/10.1007/BF00145378

Lykke, M.R., Becher, N., Haahr, T., Boedtkjer, E., Jensen, J.S. and Uldbjerg, N., 2021. Vaginal, cervical and uterine pH in women with normal and abnormal vaginal microbiota. *Pathogens*, **10**: 90. https://doi.org/10.3390/pathogens10020090

Ma, C., Du, J., Dou, Y., Chen, R., Li, Y., Zhao, L., Liu, H. and Zhang, K., 2021. The associations of genital Mycoplasmas with female infertility and adverse pregnancy outcomes: A systematic review and meta-analysis. *Reprod. Sci.*, **28**: 3013–3031.

https://doi.org/10.1007/s43032-020-00399-w

Malik, A., Rizvi, M., Jain, S., Shukla, I. and Hakim, S., 2009. P257 Association of *Chlamydia trachomatis*infection with female infertility. *Int. J. Antimicrob. Agents*, **34**. https://doi.org/10.1016/S0924-8579(09)70476-5

Mania-Pramanik, J., Kerkar, S., Sonawane, S., Mehta, P. and Salvi, V., 2012. Current *Chlamydia trachomatis* infection, a major cause of infertility. *J. Reprod. Infert.,* **13**: 204-210.

Mohseni, M., Sung, S., Takov, V., 2022. *Chlamydia*. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2022 Jan-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK537286/

Mousavi, A., Farhadifar, F., Mirnejad, R. and Ramazanzadeh, R., 2014. Detection of genital mycoplasmal infections among infertile females by multiplex PCR. *Iran J. Microbiol.*, **6**: 398-403.

Mycoplasma Infections- Statpearls- NCBI Bookshelf, 2022. https://www.ncbi.nlm.nih.gov/books/NBK536927/

Nunn, K.L., Clair, G.C., Adkins, J.N., Engbrecht, K., Fillmore, T. and Forney, L.J., 2020. Amylases in the human vagina. *mSphere*, **5**. https://doi.org/10.1128/mSphere.00943-20

Nwaziri, A.A., Ezeifeka, G.O. and Amadi, E.S., 2010. The effect of *Gardnerella vaginalis* on infertility and pregnancy of albino rats. *Int. J. Gynecol. Obstet*., **12**.

Parpillewar, M. and Singh, S., 2021. A comparative study of prevalence of *Chlamydia trachomatis* infection among infertile and fertile women at a tertiary care center. *Arch. Med. Hlth. Sci.*, **9**: 39. https://doi.org/10.4103/amhs.amhs_123_20

Peipert, J.F., Zhao, Q., Schreiber, C.A., Teal, S., Turok, D.K., Natavio, M., Cordon, S. and Daggy, J., 2021. Intrauterine device use, sexually transmitted infections, and fertility: A prospective cohort study. *Obst. Amp. Gynecol. Survey*, **76**: 746–747. https://doi.org/10.1097/01.ogx.0000806124.08178.c8

Plummer, E.L., Vodstrcil, L.A., Bodiyabadu, K., Murray, G.L., Doyle, M., Latimer, R.L., Fairley, C.K., Payne, M., Chow, E.P., Garland, S.M. and Bradshaw, C.S., 2021. Are *Mycoplasma hominis*, *Ureaplasmaurealyticum* and *Ureaplasma parvum* associated with specific genital symptoms and clinical signs in nonpregnant women? *Clin. Infect. Dis.*, **73**: 659–668. https://doi.org/10.1093/cid/ciab061

Rashidi, B.H., Chamani-Tabriz, L., Haghollahi, F., Jeddi-Tehrani, M., Naghizadeh, M.M., Shariat, M., Akhondi, M.M., Bagheri, R., Asgari, S. and Wylie, K., 2013. Effects of *Chlamydia trachomatis* infection on fertility; A case-control study. *J. Reprod. Infertil.,* **14**: 67-72.

Ravel, J., Moreno, I. and Simón, C., 2021. Bacterial vaginosis and its association with infertility, endometritis, and pelvic inflammatory disease. *Am. J. Obst. Gynecol.*, **224**: 251–257. https://doi.org/10.1016/j.ajog.2020.10.019

Salah, R.M., Allam, A.M., Magdy, A.M. and Mohamed, A.S., 2013. Bacterial vaginosis and infertility: Cause or association? *Eur. J. Obst. andamp Gynecol. Reprod. Biol.*, **167**: 59–63. https://doi.org/10.1016/j.ejogrb.2012.10.031

Sameni, F., Zadehmodarres, S., Dabiri, H., Khaledi, M. and Nezamzadeh, F., 2022. Evaluation of *Ureaplasmaurealyticum, Chlamydia trachomatis, Mycoplasma genitalium* and *Neisseria gonorrhoeae* in infertile women compared to pregnant women. *J. Obst. Gynaecol.*, **42**: 2151–2155. https://doi.org/10.1080/01443615.2022.2035328

Sharaf, M.A., El-Bohoty, S.B., Hodeib, H.A. and El-Kholi, D.G., 2021. *Chlamydia trachomatis* infection as a risk factor of infertility in women. *J. Adv. Med. med. Res.*, **39**: 63–70. https://doi.org/10.9734/jammr/2021/v33i930899

Shi, J., Dai, Y., Zhang, J., Li, X., Jia, S. and Leng, J., 2021. Pregnancy outcomes in women with infertility and coexisting endometriosis and adenomyosis after laparoscopic surgery: A long-term retrospective follow-up study. *BMC Pregnancy Childb.*, **21**. https://doi.org/10.1186/s12884-021-03851-0

Siemer, J., Larbi, Y., Theile, O., Danso, K.A., Fasching, P.A., Kreienberg, R. and Essig, A., 2008. Chlamydia trachomatis infection as a risk factor for infertility among women in Ghana, West Africa. *Am. J. Trop. Med. Hyg.*, **78**: 323–327. https://doi.org/10.4269/ajtmh.2008.78.323

Sirota, I., Zarek, S. and Segars, J., 2014. Potential influence of the microbiome on infertility and assisted reproductive technology. *Semin. Reprod. Med.*, **32**: 35–42. https://doi.org/10.1055/s-0033-1361821

Spandorfer, S.D., Neuer, A., Giraldo, P.C., Rosenwaks, Z. and Witkin, S.S., 2001. Relationship of abnormal vaginal flora, proinflammatory cytokines, and idiopathic infertility in women undergoing IVF. *J. Reprod. Med*., **46**: 806-810.

Srivastava, P., Jha, R., Bas, S., Salhan, S. and Mittal, A., 2008. In infertile women, cells from chlamydia trachomatis infected site release higher levels of interferon-gamma, interleukin-10 and tumor necrosis factor-alpha upon heat shock protein stimulation than fertile women. *Reprod. Biol.*

Endocrinol., **6**. https://doi.org/10.1186/1477-7827-6-20

Sundaram, M.K., Almutary, A.G., Alsulimani, A., Ahmad, S.R., Somvanshi, P., Bhardwaj, T., Pellicano, R., Fagoonee, S., Hussain, A. and Haque, S., 2021. Antineoplastic action of sulforaphane on HeLa cells by modulation of signaling pathways and epigenetic pathways. *Minerva Med.*, **11**.

Tantengco, O.A., de Castro Silva, M. and Velayo, C.L., 2021. The role of genital mycoplasma infection in female infertility: A systematic review and meta-analysis. *Am. J. Reprod. Immunol.*, **85**. https://doi.org/10.1111/aji.13390

Tomaiuolo, R., Veneruso, I., Cariati, F. and D'Argenio, V., 2020. Microbiota and human reproduction: The case of female infertility. *High-Throughput*, **9**: 12. https://doi.org/10.3390/ht9020012

van Oostrum, N., De Sutter, P., Meys, J. and Verstraelen, H., 2013. Risks associated with bacterial vaginosis in infertility patients: A systematic review and meta-analysis. *Hum. Reprod.*, **28**: 1809–1815. https://doi.org/10.1093/humrep/det096

Walker, M.H. and Tobler K.J., 2022. *Female infertility*. National Center for Biotechnology Information. https://pubmed.ncbi.nlm.nih.gov/32310493/

Wee, B.A., Thomas, M., Sweeney, E.L., Frentiu, F.D., Samios, M., Ravel, J., Gajer, P., Myers, G., Timms, P., Allan, J.A. and Huston, W.M., 2018. A retrospective pilot study to determine whether the reproductive tract microbiota differs between women with a history of infertility and fertile women. *Aust. N. Z. J. Obstet. Gynaecol.*, 58: 341-348. https://doi.org/10.1111/ajo.12754

Wiesenfeld, H.C., Hillier, S.L., Meyn, L.A., Amortegui, A.J. and Sweet, R.L., 2012. Subclinical pelvic inflammatory disease and infertility. *Obst. andamp Gynecol.*, **120**: 37–43. https://doi.org/10.1097/AOG.0b013e31825a6bc9

Wilson, J.D., Ralph, S.G. and Rutherford, A.J., 2002. Rates of bacterial vaginosis in women undergoing *in vitro* fertilisation for different types of infertility. *BJOG Int. J. Obst. Gynaecol.*, **109**: 714–717. https://doi.org/10.1111/j.1471-0528.2002.01297.x

Witkin, S.S., Minis, E., Athanasiou, A., Leizer, J. and Linhares, I.M., 2017. *Chlamydia trachomatis*: The persistent pathogen. *Clin. Vaccine Immunol.*, **24**. https://doi.org/10.1128/CVI.00203-17

World Health Organization, 2020. *Infertility*. World Health Organization. https://www.who.int/news-room/fact-sheets/detail/infertility