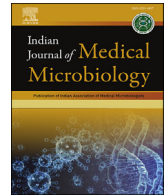




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Original Research Article

## Development of multiplex real-time quantitative PCR for simultaneous detection of *Chlamydia trachomatis*, *Mycoplasma hominis*, *Ureaplasma urealyticum*, and *Mycoplasma genitalium* in infertile women

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## ABSTRACT

**Purpose:** Sexually Transmitted Diseases (STDs) can cause sterility and many other problems for women planning pregnancy. Currently, almost 340 million people worldwide suffer from Sexually Transmitted Infections (STIs). This study made attempts to quickly identify STDs' most critical infectious agents using dedicated primers and probes.

**Methods:** The present study was done on the cervical samples of 200 infertile women. After extracting the total DNA of *Chlamydia trachomatis*, *Mycoplasma hominis*, *Ureaplasma urealyticum*, and *Mycoplasma genitalium*, quantitative methods were employed to determine the rate of target bacteria using multiplex real-time PCR.

**Results:** The multiplex qPCR showed the rates of 47%, 16%, 46%, and 16.5% for *Chlamydia trachomatis*, *Mycoplasma hominis*, *Ureaplasma urealyticum*, and *Mycoplasma genitalium* in infertile women, respectively. In some patients, there were co-infections with two or three bacteria. The diagnostic approach used in our research could be employed as an alternative detection tool to identify the four most common STD-associated bacterial agents while detecting mixed infections.

**Conclusions:** Infertile women with no biological problems could have their genital tract checked using this newly designed identification technique and get proper treatment for their infections as quickly as possible.

### 1. Introduction

Rapid molecular diagnostic methods are widely used to detect infectious agents [1,2]. The human genitourinary system is a suitable niche to grow microorganisms such as *Chlamydia trachomatis*, *Neisseria gonorrhoea*, *Streptococcus agalactiae*, *Herpes Virus type-2 (HSV-2)*, *Human papillomavirus (HPV)*, *Mycoplasma genitalium*, *Ureaplasma urealyticum*, and *Trichomonas vaginalis* [3,4]. These microorganisms are called Sexually Transmitted Infection (STI) agents. Globally, STIs are prevalent among young people and adults [5]. The organisms involved in Sexually Transmitted Diseases (STDs) can cause infection in the human genitourinary system, leading to infertility, Pelvic Infection Disease (PID), abortion, and cervicitis in women and epididymitis, urethritis, and prostatitis in men [6].

The World Health Organization (WHO) annual reports show that approximately 340 million STDs occur worldwide, with the highest rates in developing countries [7]. However, according to the WHO reports, STIs are more common in developed countries. Global statistics show that the prevalence of these infectious agents varies according to the economic situation, age, personal and community health, the number of sexual partners, and the community's social conditions. Therefore, STIs are generally considered a major global problem with devastating outcomes, including financial loss and family damage [8].

There are no signs of STDs in the presence of STI agents, a situation that fails correct diagnosis and treatment of STIs [9]. Detecting *C. trachomatis* is essential when looking for STI agents because it causes infection in the genitourinary system. Many studies have shown that *C. trachomatis* can be a co-factor for Cervical Intraepithelial Neoplasia (CIN)

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