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Review Article

Oncology Section

# Association of Genital Infections Other Than Human Papillomavirus with Pre-Invasive and Invasive Cervical Neoplasia

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## ABSTRACT

Human papillomavirus (HPV) is a well-established causative agent of malignancy of the female genital tract and a common Sexually Transmitted Infection. The probable co-factors that prevent spontaneous clearance of HPV and progression to neoplasia are genital tract infections from organisms like *Chlamydia*, *Trichomonas vaginalis* etc, smoking, nutritional deficiencies and multiparity. Inflammatory conditions can lead to pre-neoplastic manifestations in the cervical epithelium; however their specific role in cervical carcinogenesis is not yet established. Therefore it is imperative to study the likely association between HPV and co-infection with various common pathogens in the genital tract of women having cervical precancer or cancer. A "Pubmed" search was made for articles in Literature on this topic using the words: Cervical neoplasia, HPV, co-infections, Cervical Intraepithelial Neoplasia (CIN), *Trichomonas vaginalis*, *Candida*, *Chlamydia* and the relevant information obtained was used to draft the review.


**Keywords:** *Candida, Chlamydia*, Co-infections, *Trichomonas vaginalis*

## INTRODUCTION

Infection from high risk types of Human Papilloma Virus (HPV) is an important but not the sole cause of cervical cancer [1]. Persons belonging to adolescent age group and having history of exposure are prone to infection with HPV, the risk is increased by tobacco smoking, oral contraceptive use, poverty, simultaneous infection with other sexually transmitted organism, persistent and recurrent inflammation, HIV or other infection that depresses the immune system [2]. These agents are co-factors in pathogenesis of cervical cancer, HPV transmissibility, persistency, progression and HPV induced carcinogenesis. Besides HIV and HPV, *Chlamydia trachomatis*, *Trichomonas vaginalis* and *Candida* spp. are other sexually transmitted pathogens associated with cervical neoplasias. Recurrent inflammation facilitates cellular proliferation and shedding of the epithelium, and helps in growth of malignant clone of cells; cytokines, chemokines, free radicals and growth factors help colonization of microbes [3]. Cancers occur when cells with integrated viral genetic material escape normal cell cycle control mechanisms [4].

This article is a review of literature regarding the prevalence of different sexually transmitted organisms considered as co-infections, pathophysiology of diseases caused by them and the type and extent of their association with pre-invasive and invasive cervical cancer. A brief outline of studies of last fifteen years included in the review is given in [Table/Fig-1] [5-34].

## CO-INFECTIONS AND CERVICAL NEOPLASIA

### 1. *Candida*

#### a. Nature & mechanism of transmission of pathogen

Fungi from *Candida* genus are human commensal flora and may be isolated from genital organs. Person to person transmission of this fungal infection is common and so genital *candida* infection is seen in sexually active age group. Vulvo-vaginal candidiasis is most often caused by *Candida* albicans although other species can also be identified.

#### b. Prevalence of general population & in patients with CIN and Invasive Cancer

Prevalence of *Candida* spp. by microscopy and culture has been found to be 20% in patients without any abnormality detected on colposcopy and 11% in premenopausal patients with abnormal colposcopy [35]. In Nigeria, *Candida* spp. was obtained on cytological examination in 2.2% women undergoing community screening programme. [36], in 6.7% in a cervical cancer screening programme in Iran [27] and 12% by cytology in China [29]. Vaginal *Candida* species colonization alone was not found to be significantly associated with all the high risk HPV genotypes (OR=0.45, 95% CI: 0.23-0.87) [23], women carrying *Candida* spp. were not found to be at an increased risk of developing cervical cancer [13]. Risk of development of cervical neoplasia was not increased by vaginal fungal infection in HPV positive or negative subjects [37].

#### c. Prevalence of *Candida* in India

*Candida* spp. was detected in vaginal samples by cytology in 0.8% women having normal smears in Delhi [10]. Microscopy and culture detected Candidiasis in 10.33% among female sex workers (n=300) as reported from a study in Surat [38], in 90% of HPV negative high risk group comprising of FSWs in West Bengal and 88.6% in HPV positive FSWs in the same region (p=0.69) [25]. A cytology based study in West Bengal had failed to establish any significant association between *Candida* infection and cervical dysplasia [39].

There are no reports in literature establishing conclusively that fungal infection of vagina is associated with higher incidence of cervical neoplasia either in the presence or absence of HPV, no marked difference in prevalence rate was observed between different geographical areas. Studies employing culture technique, observed a higher positivity than those relying on smear examination alone. The detection of *Candida* spp. in dysplastic lesions of the cervix does not prove a cause and effect association.

#### d. Association with HPV & genital cancer; possible role in carcinogenesis

*Candida* species comprise the commonest fungal infection of the genital tract, in many cases only asymptomatic colonization takes place; however some highly pathogenic and virulent species of *Candida* exist which cause protein degradation and enhance antigenic response leading to mucosal injury and endogenous invasion [33]. The extent of damage incurred is also determined by inherent property of epithelial cells like state of maturity [40]. In a person harbouring vulvo vaginal candidiasis, the infectious milieu

| S. No. | Author | Number recruited | Co-infections studied | Verification of disease status (HPV test by PCR, Oncogenic HPV test; HPV by cytology; SIL on cytology; colposcopy; histology) |
|---|---|---|---|---|
| 1. | Gopalkrishna V, 2000 (India), case control [5]. | n = 80 | Herpes Simplex virus<br>*Candida*<br>Bacterial vaginosis<br>*Chlamydia*<br>Gonorrhoea<br>Syphilis | HPV test by PCR<br>Oncogenic HPV test by PCR. |
| 2. | Antila T, 2001 (Finland), Longitudinal nested case control [6]. | n =Cohort of 530000<br>SCC=128<br>Control=3 matched control for each case | *Chlamydia* | Histology<br>HPV by Serology |
| 3. | Wallin KL, 2002 (Sweden), Case control, Prospective [7]. | N = 236 | *Chlamydia* | SIL on cytology<br>Histology<br>HPV test by PCR |
| 4. | Tamim H, 2002 (Lebanon), Case control [8]. | n = 129<br>HPV DNA –ve=80<br>HPV DNA +ve= 40 | *Chlamydia* | SIL on cytology<br>HPV test by PCR |
| 5. | Castle PE, 2003 (Jamaica) [9]. | n = 447 | Herpes Simplex virus<br>*Chlamydia*<br>Human T Cell<br>Lymphotrophic Virus<br>Type 1(HTLV-1) | Colposcopy |
| 6. | Madhu J et al., 2004 (India), case control, cross Sectional [10]. | n =1308<br>No CIN=1024<br>CIN=284<br>Low grade CIN=204<br>High grade CIN=80 | Herpes Simplex virus<br>*Trichomonas vaginalis*<br>*Candida*<br>Bacterial vaginosis<br>*Chlamydia*<br>Syphilis<br>HIV | HPV by cytology<br>SIL on cytology |
| 7. | Smith JS, 2004 (Thailand, the Philippines, Morocco, Peru, Brazil, Colombia and Spain, coordinated by the International Agency for Research on Cancer, Lyon, France), Case control [11]. | n = 1238<br>Control=100 | *Chlamydia* | *Chlamydia* antibody by micro-immunofluorescence<br>HPV DNA |
| 8. | Samoff E, 2005 (Atlanta), longitudinal, Cohort [12]. | n = 621 | *Trichomonas vaginalis*<br>Bacterial vaginosis<br>*Chlamydia*<br>Gonorrhoea | SIL on cytology<br>HPV test by PCR<br>Oncogenic HPV test by PCR and sequencing. |
| 9. | Naucler P, 2007 (Taiwan), Prospective cohort followed by nested case control [13]. | n = 13595<br>Cases= 114 SCC,<br>Control=519 | *Chlamydia* | HPV by Serology<br>SIL on cytology<br>Histology |
| 10. | Madeleine MM, 2007 (Seattle), Case control [14]. | n = 805<br>SCC=302<br>AC-185<br>HPV + Control=318 | *Chlamydia* | Histology |
| 11. | Zereu M, 2007 (Brazil) [15]. | n = 206 Cervical Adenocarcinoma specimen | Herpes Simplex virus<br>*Chlamydia* | Histology<br>HPV test by DNA Sequencing |
| 12. | Chernesky M, 2007 (Canada) [16]. | n=290 | *Chlamydia* trachomatis,<br>Neisseria gonorrhoea | NAAT for rRNA |
| 13. | Engberts MK, 2008, (Netherlands), Cohort [17]. | n = 445,671 | *Candida* | SIL on cytology |
| 14. | Quint KD, 2009 (Netherland) [18]. | n = 71 Cervical Adenocarcinoma specimen | *Chlamydia* | Histology<br>HPV test by PCR<br>Oncogenic HPV test by Genotyping |
| 15. | Safaeian M, 2010 (Rockville), Prospective cohort followed by nested case control [19]. | n = 10049, out of which CIN=314,<br>Control=995 | *Chlamydia* | Histology<br>HPV test by PCR and dot-blot hybridization. |
| 16. | Valadan M, 2010 (Middle East), Case control [20]. | n = 145 | *Chlamydia* | Colposcopy<br>Histology |
| 17. | Krashin JW, 2010 [21]. | n = 467 | *Trichomonas vaginalis* | Culture |
| 18. | Farivar TN, 2012 (Tehran), Case control [22]. | n = 226<br>(Cases-76<br>Control-150) | *Chlamydia* | Histology |
| 19. | Rodriguez-Cerdeira, 2012(Spain), Cross-sectional [23]. | n = 208 | *Trichomonas vaginalis*<br>*Candida*<br>Bacterial vaginosis | Oncogenic HPV test by HC2 |
| 20. | Ginocchio CC, 2012 (United States) [24]. | n = 7593 | *Trichomonas vaginalis*<br>*Chlamydia trachomatis*, Neisseria gonorrhoea | NAAT for rRNA |

| S. No. | Author | Number recruited | Co-infections studied | Verification of disease status (HPV test by PCR, Oncogenic HPV test; HPV by cytology; SIL on cytology; colposcopy; histology) |
|---|---|---|---|---|
| 21. | Ghosh et al., 2012 (India), case control, cross-Sectional [25]. | n = 45<br>HPV +ve = 35<br>HPV-VE = 10 | Herpes Simplex virus<br>*Trichomonas vaginalis*<br>*Candida*<br>*Chlamydia*<br>Syphilis<br>HIV | HPV test by PCR<br>Oncogenic HPV test by PCR<br>SIL on cytology |
| 22. | Silva J et al., 2013 (Southern Europe) [26]. | n = 432 | *Chlamydia trachomatis*, HPV | PCR |
| 23. | Kalantari N, 2014 (Iran), Retrospective [27]. | n = 33600 | *Trichomonas vaginalis*, *Candida* spp., Gardenella vaginalis | Cytology |
| 24. | Bellaminutti S, 2014 (Italy), Cohort [28]. | n = 441 | *Chlamydia trachomatis*, HPV | Bead based molecular technique |
| 25. | Zhou H, 2014 (China), Cohort [29]. | n = 46866 | *Trichomonas vaginalis*, *Candida* spp., HPV | Cytology |
| 26. | Saleh AM, 2014 (Sudan), cross-sectional [30]. | n = 297 | *Trichomonas vaginalis* | Latex agglutination, culture, PCR |
| 27. | Swartzendruber A, 2014 (Atlanta) [31]. | n = 605 | *Trichomonas vaginalis*<br>*Chlamydia trachomatis*, Neisseria gonorrhoea | CT NG Amplified DNA Assay, PCR for TV |
| 28. | Chernesky M, 2014 (Canada) [32]. | n = 708 | *Trichomonas vaginalis*, *Chlamydia* trachomatis, | NAAT for rRNA |
| 29. | Jensen KE, 2014 (Denmark) [33]. | n = 1390 | *Chlamydia trachomatis*, HPV | HPV and CT DNA |
| 30. | Gunasekera HAKM, 2014 [34]. | n = 168 | *Chlamydia trachomatis* | DNA Amplification and Hybrid Capture |

**[Table/Fig-1]:** List of reports showing co-infections studied and methodology used

serves as a gateway for infection with other organisms including Human papillomavirus facilitating its entry and propagation. In endogenous fungal infection, tissue debris and accumulation of free-radicals enhances virulence of the organism and increases susceptibility of the host [41]. Therefore, the association of *Candida* spp. with cervical lesions may be related to its inflammatory effects.

## 2. *Trichomonas vaginalis*

### a. Nature & mechanism of transmission of pathogen

One of the commonest cause of vaginal discharge is infection by the sexually transmitted parasite *Trichomonas vaginalis* belonging to the family "protozoa", characterized by rippling motility in wet mount of genital samples collected from cases with leucorrhea, reported first by Donnè; the obligatory parasitic nature of the organism is because of its dependency on host secretions and tissue debris for nutritious elements containing nucleic acids and lipids [42]. The infection manifests by malodorous, frothy vaginal discharge, redness, swelling and punctuate hemorrhagic spots characteristically described as "colpitis macularis or strawberry cervix" [42,43]. Infection of neighbouring organs like urethra, urinary bladder and endocervical canal might also occur in addition to vaginitis which is the predominant symptom [44].

### b. Prevalence of general population and in patients with CA or CIN

According to a report from Centre for Disease Control, 14% of HIV negative females who were sexually active were infected with *Trichomonas vaginalis* [21]. Cytology based screening programmes conducted in Nigeria, reported prevalence of *Trichomonas vaginalis* of 2.52% [36], an Iranian study found 0.2% prevalence rate [45], a Chinese study reported 4.0% in China while positivity of 8.7% was found by a NAAT based rRNA detection technique in the United States [24]. Patients with normal colposcopic findings had Trichomonas infection rate of 2% by microscopy and culture while the prevalence of infection was 3% among the total cases screened [35].

The genotypes of HPV found to be associated with *Trichomonas vaginalis* included high risk types 18, 45, 66 and 68 [23]. A meta-analysis including 24 studies had been performed by Zhang and Begg and it was concluded that a significant association existed between TV infection and cervical neoplasia (including both squamous intraepithelial lesions and cervical neoplasia) [45]. A similar observation that Relative Risk of acquiring pre-neoplastic and neoplastic lesions of the cervix was higher in persons infected with *Trichomonas vaginalis* had been made by Vikki et al., in a prospective study (Standardized Incidence Ratio 6.4) [37]. The temporal co-relation between high grade cervical intraepithelial neoplasia or cancer with HPV and *Trichomonas vaginalis* infection had been analysed by Gram IT et al., who concluded that these infections played a predominant role in disease pathogenesis [46]. A high prevalence rate of TV had been obtained by Jatau ED et al., by wet mount and culture (overall prevalence of TV among antenatal cases 18.7%) [47]. Lazenby GB et al., had found TV by Nucleic acid amplification test (NAAT) to be associated with an increased risk of acquisition of high risk HPV (OR 4.2, 95% C.I. 1.7-10.3) [48]. Some other authors have also reported a high prevalence of Trichomoniasis in female patients (Zigas et al., 44.6% among STD clinic attendees [49], O'Farrell et al., 49.2% in antenatal clinics by culture method [50], Swartzendruber A et al., 17% in a reproductive health clinic among African American adolescents by PCR [31]. In cytology based study conducted separately in rural and urban settings, prevalence of *Trichomonas vaginalis* was 27.4% and 29.6% respectively [36] while in another rural population based study, prevalence was found to be 24.7% [51]. Passey et al., reported positivity of 46% by wet mount in a community based random cluster sampling [52]. Saleh AM et al., conducted a study on 297 women in Sudan with vaginal discharge and obtained varying prevalence of Trichomoniasis, though within a close range of 84.8% to 86.5% after comparing different techniques of detection like wet mount, latex agglutination, culture and molecular methods [30]. Isolation rates of *Trichomonas vaginalis* is influenced strongly by socio-demographic parameters of the study groups like residence, religion, age, sex; population recruited that is STD clinic attendees or women undergoing routine screening in community, clinical presentation defined by presence or absence of symptoms and laboratory methodology used [53,54].

### c. Prevalence of *Trichomonas vaginalis* in India

The prevalence of trichomoniasis from different parts of India ranges from 0.4-27.4% according to published literature [42]. A cytology based study from North India had observed 7.8% prevalence

of trichomoniasis among normal population, 4.2% in cases with cervical intraepithelial lesions and 5.9% in cases with low grade CIN [10]. Prevalence of *Trichomonas vaginalis* was 10% among women with vaginal discharge attending a reproductive health clinic in the same region [55]. The prevalence of Trichomoniasis by culture, had been found to be 22.9% among HPV positive FSWs in West Bengal which was significantly higher than cases with no HPV infection (p=0.04) [25].

Therefore observations of cross-sectional as well as longitudinal follow up studies in India as well as in the West demonstrate significant association of *Trichomonas vaginalis* with pre-invasive and invasive cervical lesions, indicating that Trichomoniasis is a predictor of cervical neoplasia. An increase in isolation rate was obtained by studies employing NAAT or culture compared to those where detection was by wet mount or cytology.

### d. Association with HPV & genital cancer; possible role in carcinogenesis

The association of *Trichomonas vaginalis* with high grade cervical intraepithelial lesions is because of the epithelial alteration and damage that characterize these conditions facilitate proliferation of the organism [56]. The cell mediated immunity generated against the invading organism involves recruitment of leucocytes in large numbers which are associated findings on cytology of smears, further the parasite often imbibes nutritious elements like fatty acids and iron by destruction of host red blood cells which is caused by cytotoxic trypsin like substances called cell detaching factor, CDF and N-nitrosamines liberated during infection, which also promote the process of epithelial atypia and dysplasia [42]. Vaginal pH rises during Trichomonas infection, which is condusive to growth of the organism. *Trichomonas vaginalis* thrives on tissue debris and serous exudate and produces tissue damage that are extensive and atypical. The presence of the organism has a high incidence of association with both systemic and local conditions of great clinical significance [44]. Disease pathogenesis of *Trichomonas vaginalis* infection necessitates multitude of cross-communications involving viruses, bacteria, eukaryotes and human host [57].

## 3. *Chlamydia trachomatis*

### a. Nature & mechanism of transmission of pathogen

*Chlamydia trachomatis* is a gram negative bacterium that only grows inside cells [58]. The life cycle is complex and is divided into two parts: the reticulate body which divides to form the elementary body which is the infectious agent [59].

The organism causes male and female lower genital tract infection, such as non-gonococcal urethritis and cervicitis; it may cause pelvic inflammatory disease (PID) and endometritis (serovar D to K). Three specific serovars (L1 to L3) cause lymphogranuloma venereum, other serovars (A to C) cause trachoma and inclusion conjunctivitis [60]. The mode of transmission is by sexual intercourse [58].

### b. Prevalence of general population & in patients with CA or CIN

*Chlamydia trachomatis* infection comprises a predominant sexually transmitted infection in the United States. Overall positivity of *Chlamydia trachomatis* was found to be 8.7% among young American women where the prevalence demonstrated inverse relationship with age; the highest prevalence was noted in age group of 15-19 years (15.3%-18.6%) and lowest in cases older than 35 years (1.0%-2.6%) [61], Swartzendruber A et al., found 21% positivity by DNA amplification among African-American individuals belonging to adolescent age group [51]. A prevalence of 8.3% positivity had been noted among STD clinic attendees using DNA Amplification and Hybrid Capture [34]. Ginocchio CC et al., found 6.7% positivity by NAAT based rRNA detection technique [24]. *Chlamydia trachomatis* was isolated in 18% of cases with inflammatory cervical smears of which, 8% occurred in patients having normal colposcopy and 33% among cases having colposcopic abnormality [35].

An increased incidence of cervical squamous cell carcinoma occurs in persons infected with *Chlamydia trachomatis* [11]. HPV positive women are more prone to acquire *Chlamydia trachomatis* infection than HPV negative women [7,8,62]. The risk of cervical carcinogenesis was found to be higher in women exposed to multiple and specific serotypes [6,14,63]. Serum antibodies to *C trachomatis* were associated with enhanced risk for acquiring CIN [20,64-66]. The prevalence of *Chlamydia trachomatis* had been found to vary on use of different methods of detection. By PCR and immuno-fluorescence method, Bulhak-Kozioł V et al., had found *Chlamydia trachomatis* positivity of 12.2-20% in cases with cervicitis or erosion and 27.8-34% positivity by serological techniques like IgG ELISA [67]. Dicker LW et al., applying Transcription Mediated Amplified (TMA) DNA Assay, found 8.5% positive rate of *Chlamydia*l infection [61], Bellaminutti S et al., reported 9.7% positivity in a cervical cancer screening programme by molecular methods employing coated beads [28], Chernesky M et al., found positivity of 10% in Canada based on L-PAP samples, they found varying sensitivity and specificities on comparing kits from different manufacturers [16] and established strong agreement between results obtained on self collected and physician collected samples [32].

On the other hand, several studies in literature have failed to demonstrate significant association between Clamydia and cervical preneoplasia and cancer [9,13,15,18,19,22,33,68]. Lazenby GB et al., had not found any case of *Chlamydia trachomatis* using the technique of rRNA detection by TMA [48]. It has been suggested that a causal association exists between HPV and *Chlamydia trachomatis* in young women, CT may predispose subsequent acquisition of HPV infection and development of cervical neoplasia [26].

### c. Prevalence of *Chlamydia trachomatis* in India

Bhatla N et al., had found CT DNA by Hybrid Capture assay to be prevalent in 4.8% Indian women and HPV/CT co-infection in 0.7% [69]. *Chlamydia* was detected cytologically in 0.92% of total women screened among which 0.4% were among cases free from CIN, 2.8% women with CIN, of which 10% occurred in women suffering from high grade CIN [10]. By direct antigen detection test, *Chlamydia* prevalence was found to be 20% among HPV negative vs. 14% in HPV positive female sex workers however the discrepancy did not reach the level of statistical significance (p=0.8) [25]. Among attendees of a Delhi based reproductive health clinic, *Chlamydia trachomatis* positivity was 12.2% in women complaining of vaginal discharge [55]. By PCR technique, positivity of *Chlamydia* was found to be 12-22% [5]. A study from West Bengal had found association between cytological evidence of *Chlamydia trachomatis* and cervical dysplasia [39].

The prevalence of *Chlamydia*sis ranged from 0.9%–20% in the various studies, heterogeneity of opinion was noted as several studies had demonstrated strong association of genital *chlamydia*sis with persistence of high risk HPV and cervical squamous cell carcinoma that may not be serotype specific whereas lack of similar association was also reported by many authors. Overall the prevalence of *Chlamydia* in India was lower compared to that reported in Western Literature. *Chlamydia trachomatis* infections are most commonly detected in women less than 25 years of age [67]. The highest prevalence of *Trichomonas vaginalis* has been found in women >40 years of age while *Chlamydia trachomatis* prevalence is lowest in that age group [24]. The recommended age for cervical cancer screening based on high risk HPV detection tests is30-65 years [70-72]. This could be one of the reasons for emergence of greater association between Trichomonas than *Chlamydia trachomatis* with HPV associated cervical neoplasias.

### d. Association with HPV & genital cancer; possible role in carcinogenesis

Epithelial cells infected with *Chlamydia trachomatis* become susceptible to infection with high-risk Human papilloma virus and the synergistic actions of the two infectious organisms leads to development of neoplasia [64]. In persistent and recurrent *Chlamydia*l infection, liberation of cytotoxic substances like nitric oxide as well as anti-apoptotic mechanisms come into play resulting in proliferation of damaged cells and initiating carcinogenesis, the co-factor role of the organism in HPV associated cervical lesions can be attributed to immune-modulation [8]. As a result of the disturbances and under the influence of persistent infection, the cells escape the control of the cell signalling mechanisms, DNA damage occurs leading to proliferation of clones of cells carrying altered genetic material with enhanced propensity for neoplastic change [73].

The sites of infection by *Chlamydia trachomatis* are columnar epithelial cells of the endocervix as is evident by increased prevalence of infection in cases with cervical ectropion; regions of squamous metaplasia of cervix are increasingly infected by *Chlamydia* accounting for the high prevalence of squamous cell carcinoma in association with the infection [64,74]. When a cell is infected with *Chlamydia trachomatis*, entry of HPV to the basal layer is facilitated by microscopic epithelial injuries, HPV viral particles accumulate and derangement of host immunity occurs which is manifested by shift of immune response from T-helper cell type 1(active in HPV control) to T-helper cell type 2 and plasma cell infiltrates [12].

### Observations on Reported Studies

Many of the studies in literature had diagnosed co-infections of HPV on cytology. Cytological interpretation of infections may not always be fully accurate. Studies that rely on cytological assessment of HPV have their limitations [46]. Variability in results are observed depending on the diagnostic techniques employed like wet-mount, culture, immunochromatographic techniques, serology, PCR, NAAT which have different sensitivity and specificity. Another drawback is that the number of recruits in many studies was small and observation spanned over a limited time period. Evaluation of larger cohort of subjects over a prolonged time frame is required to assess the influence of co infections like *Trichomonas vaginalis*, *Chlamydia trachomatis* and *Candida* spp. on HPV pathogenesis and vice versa and to study the combined role and modes of action and interaction of these organisms in development of malignant conditions.

## CONCLUSION

In summary, there is no definite indication that infection with Candidia spp. enhances the risk for cervical carcinogenesis. *Trichomonas vaginalis* infection is an important risk associate of cervical malignancy singly and associated with HPV. There is heterogenecity of data regarding causal association of *Chlamydia* with cervical cancer; reports differ according to population studied. Routine screening programmes often detect infection by multiple sexually transmitted organisms. It is important to screen for genital infections, particularly in HPV positive patients to identify the presence of other microorganisms to reduce the probable cumulative effects of vaginal flora in promoting HPV persistence and cervical carcinogenesis.

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