

Study of Concomitant Fungal, Bacterial, Viral, and Parasitic Infections with HPV in Brazilian Women

Tangará Jorge Mutran^{1*}, Ismael Dale Cotrim Guerreiro da Silva² & Silvia M. Fernandes Brenna³

^{1*} Teacher Coordinator of Biomedicine of Anhembi Morumbi University, São Paulo SP. Brazil.

² Research Coordinator of Department of molecular Gynecology, Federal University of São Paulo. SP Brazil.

³ Student of Sao Paulo City University, School of Medicine. Sao Paulo, SP Brazil.

Accepted 20th April, 2015

Abstract

Background: Studies have shown that human papillomavirus HPV screening is protective against the development of cervical cancer. Early initiation of sexual activities, the number of sexual partners, smoking, and according to Zur Hausen, the presence of "mutagenic" infections, are all risky factors for the development of cervical cancer. **Methods:** This investigation consisted of descriptive observation of 200 sexually active women 18-30 years old, taking into account the mutagenic frequency of several infections in the general population. **Results:** The infections incidences in the study population were: HPV, 13.5%; *Staphylococcus* spp., 46.5%; *Streptococcus* spp., 4.0%; *Trichomonas vaginalis*, 4.5%; genus *Candida*, 25.5%; herpes simplex, 0%; and *Chlamydia trachomatis*, 5%. **Conclusions:** Fisher's Chi-square test was applied to socioeconomic data and infections data, but no significant associations were uncovered between the presence of HPV and any of the factors listed.

Keywords: infection, HPV, cervical cancer

Introduction

Cervical cancer is the second most common cancer among women worldwide in the study by Parkin¹, with approximately 500,0 new cases per year and approximately 230,0 deaths yearly, as per the Brazilian National Cancer Institute². In Brazil, cervical cancer is the third most common cancer in the female population, as reported by the National Cancer Institute². In 2010, 18 new cases are estimated to have occurred per 100,0 women. Excluding non-melanoma skin tumors, the uterine cancer is the most prevalent cancer in the north of Brazil 23/100,0, the second most common in the midwest 20/100,0 and the northeast 18/100,0, the third most prevalent cancer in the south 21/100,0 and the southeast of Brazil² 16/100,0.

Although human papillomavirus HPV infection is an important factor in the development of cervical cancer, it does not act alone. The importance of non-viral factors associated with cervical cancer has been stressed; these factors include smoking, high parity, high number of sexual partners, premature initiation of sexual activity, and the concomitant presence of lower genital tract infections. In 2002, Zur Hausen³ coined the term "mutagenic infections" to describe infections induced by bacteria, protozoa, and viruses. In this context, previous investigations have sought

co-factors associated with HPV infection that, at least, provide evidence of latent infection, test of Focaccia⁴. Despite the extensive knowledge of the microbiology of normal and abnormal flora linked to HPV, many issues have not been fully clarified. Therefore, in the present article, our aim was to evaluate the frequencies of concomitant fungal, bacterial, viral, and parasitic infections with HPV and its socioeconomic issues in a group of sexually active women.

Objective

To assess the frequency of concomitant fungal, bacterial, viral and parasitic infections with HPV and its socioeconomic issues in a group of sexually active women.

Materials and Methods

A cross-sectional observational study was performed which included 200 sexually active women between 18 and 30 years old who exhibited appropriate physical and psychological conditions for a material vaginal collection. The studied subjects were asymptomatic women who had chosen to undergo routine testing without physician referral. All subjects had read and signed an informed consent form. The office visiting occurred at the basic health units in the São Paulo and Mairiporã municipalities, Brazil. *Trichomonas vaginalis* was assayed through the technique used by Ohlemeyer⁵ and co-workers, which was described as the highest specificity test, 99,8%, by Jitraphai and Wise⁶.

Detection of *Streptococcus* spp. and *Staphylococcus* spp. was carried out according to the manual of clinical microbiology of infection control in health care recommendations⁷. The methodology used for the detection of *Candida* spp. and *Candida albicans* was described in Neto⁸ and colleagues, and the identification was made through germ tubes test by Hinrichsen⁹.

For the diagnostics of HPV, *Chlamydia trachomatis*, and the herpes simplex, we performed molecular analysis by polymerase chain reaction PCR as described by Mahony¹⁰ and co-workers.

The primers used in the identification for the sexual infections studied is to *C. trachomatis* 5' CGC TTG GGA ATA

Corresponding Author: Tangará Jorge Mutran

Teacher Coordinator of Biomedicine of Anhembi Morumbi University, São Paulo SP

E-mail: jolive17@outlook.com

AGA GAA GAC 3' (Forward) 5' CGC TTG GGA ATA AGA GAA GAC 3' (Reverse), for HPV 5' TTT GTT ACT GTG GTA GAT ACT AC 3' (GP5). 5' GAA AAA TAA ACT GTA AAT CAT ATT C 3' (GP6). 5' CGT CCM ARR GGA WAC TGA TC 3' (MY 9) and 5' GCM CAG GGW CAT AAY AAT GG 3' (MY 11). For the *Herpes simplex* identification using primers pair 5' CAG TAC GGC CCC GAG TTC GTG ACC GGG 3' (Forward) and 5' GGC GTA GTA GGC GGG GAT GTC GCG 3' (Reverse)

Results

Figure 1 shows that among the studied women, 13.5% had HPV infection. Moreover, after genotyping by PCR-RFLP, we verified that among the 27 positive samples for HPV, genotype 16 predominated in 26% of cases. Furthermore, in the positive samples the following agents: *Candida sp*, *Candida albicans*, *Streptococcus*, *Staphylococcus*,

trichomonas, HPV, *Chlamydia trachomatis*, and *herpes simplex*. However, the most common infectious agent, *Staphylococcus spp*, was present in 45% of the samples, followed by *Candida spp* 30%, *Candida Albicans* 25%, *Chlamydia* and HPV 15%. The percentage of samples with positive HPV infection and gram-positive cocci were the most common (Figure 2). The white race was the one that presented a higher number of positive results in almost agents tested, except for *Candida* and *Chlamydia* (Figure 3).

People with lower income presented a higher number of positive results for all the tested agents (Figure 4). The person that does not work had presented a higher number of positive results for all the tested agents (Figure 5). The married women are the ones that had presented the highest number of negative results for all the tested agents (Figure 6).

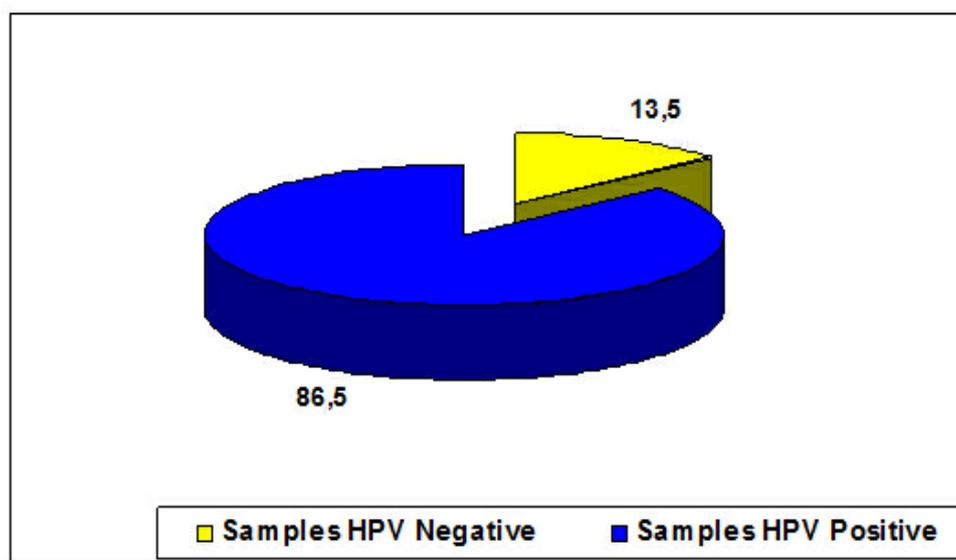


Figure 1. Positive diagnosis for HPV was generated for 13.5% of the samples.

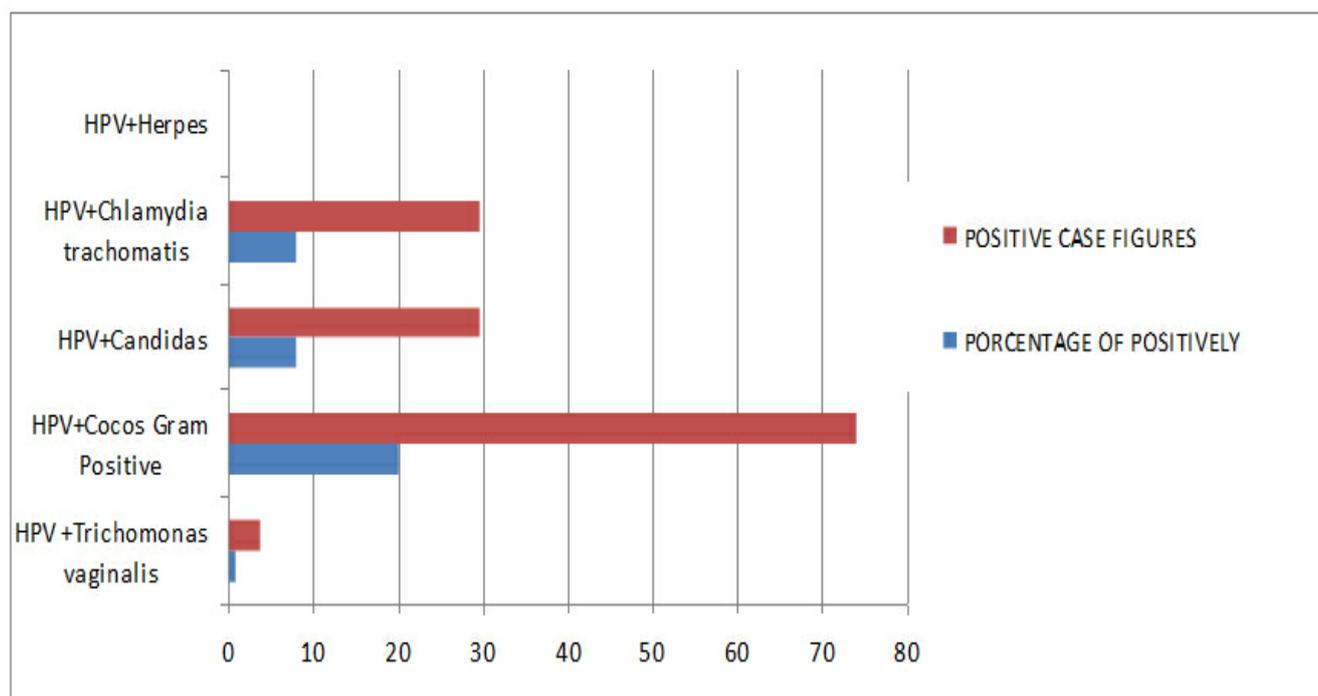


Figure 2. Percentage of HPV-positive samples positive for concomitant infections.

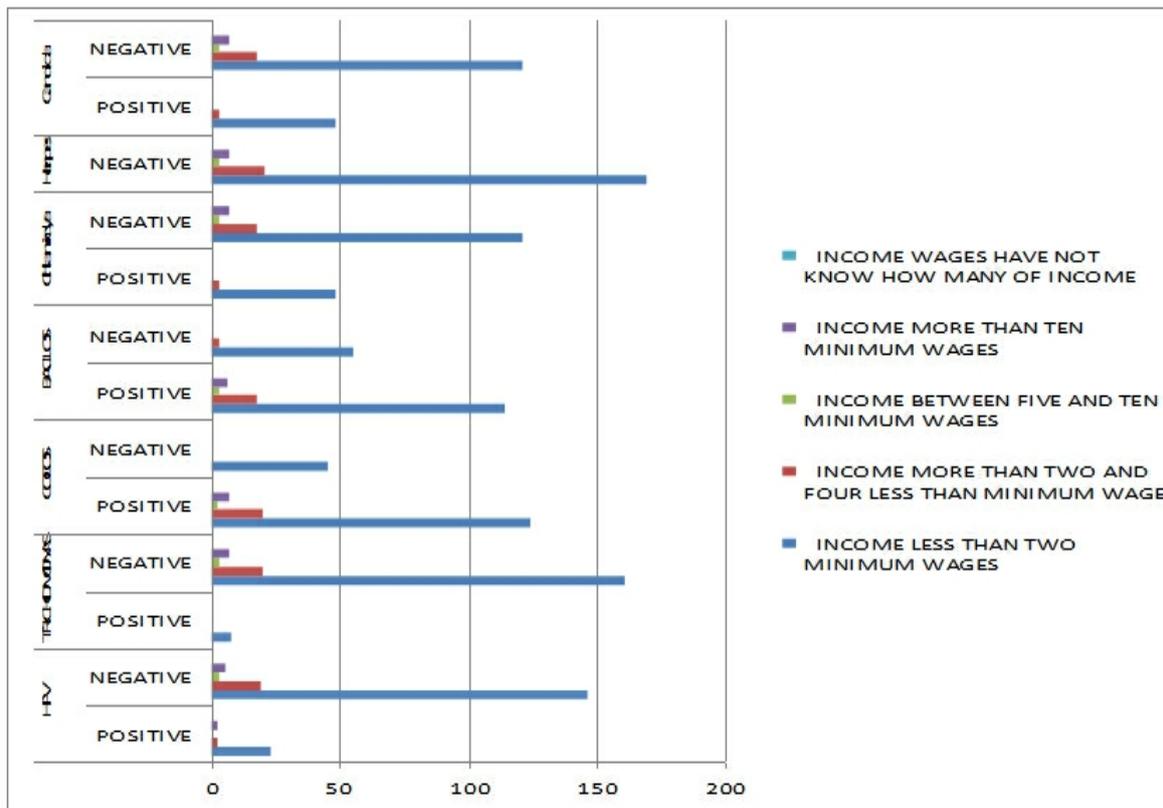


Figure 4. The relationship between income and infections.

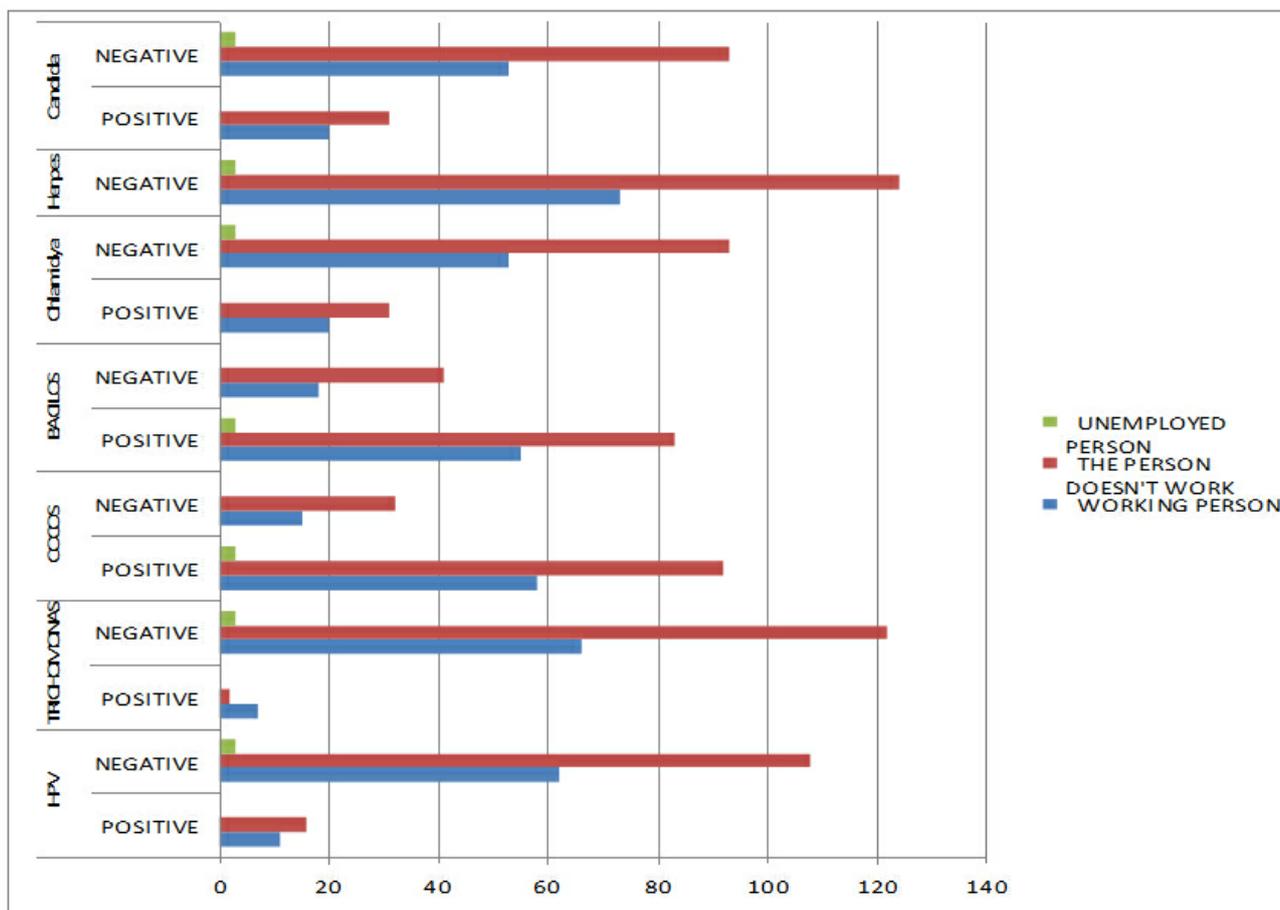


Figure 5. The relationship between occupation and infection.

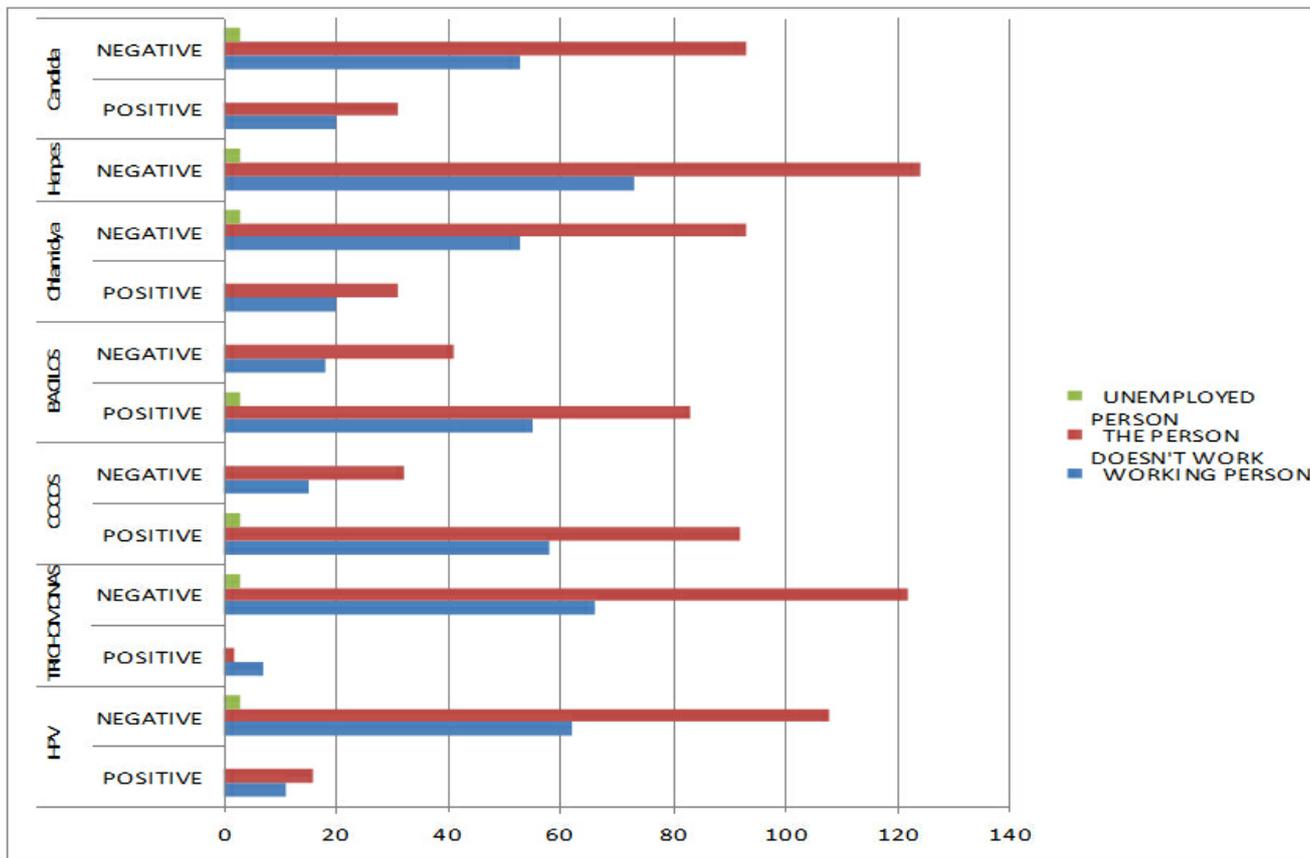


Figure 5. The relationship between occupation and infection.

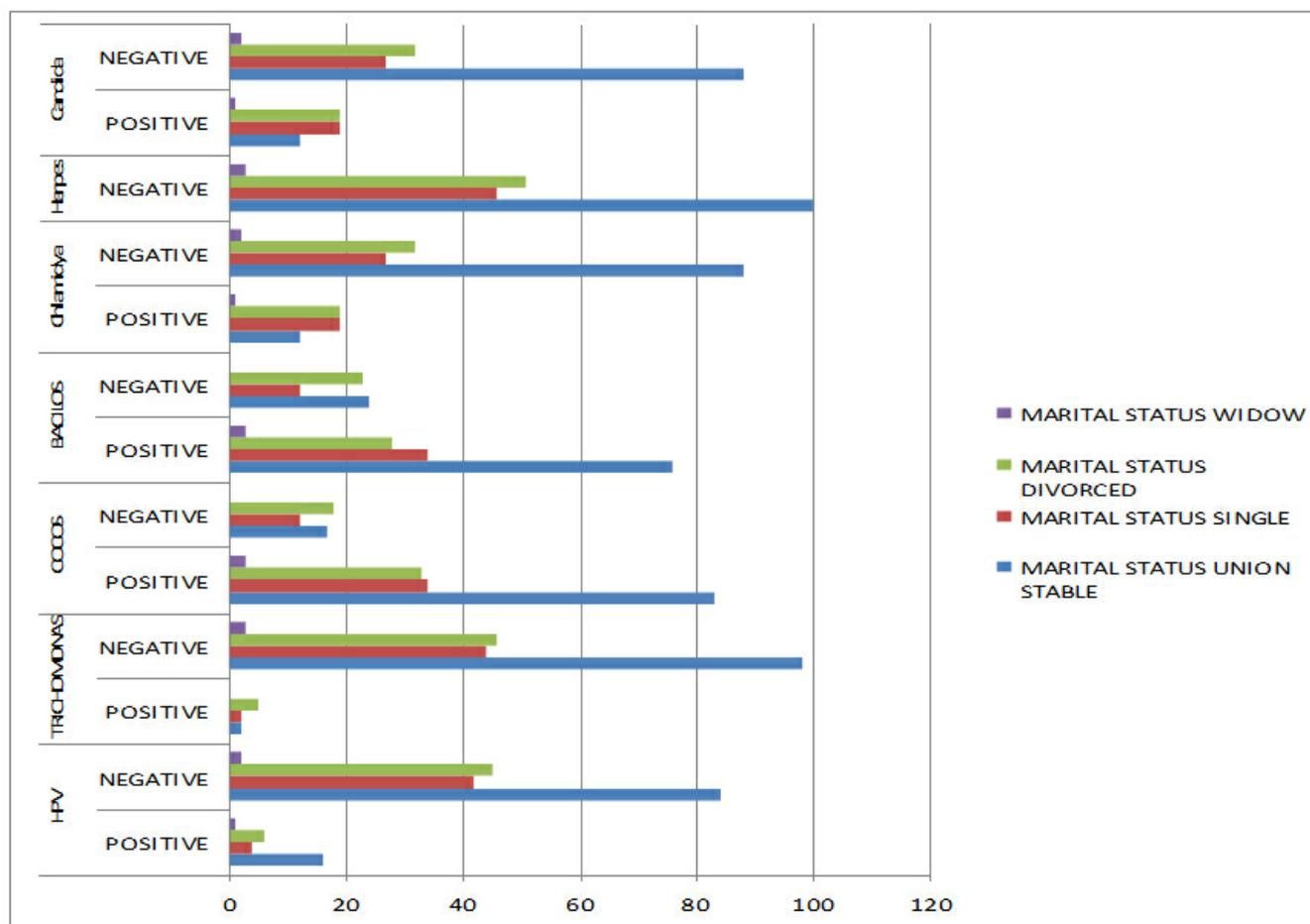


Figure 6. The relationship between marital status and infections.

Discussion

The present study was assessed the concomitant presence of bacterial, viral, fungal, parasitic infectious agents and psychosocial data in women with HPV in the city of São Paulo.

We included sexually active women between 20 and 30 years old who pursued spontaneously and voluntarily the health care during the campaigns called "Prevention of the Cervix Cancer", to perform the pap smear exam, obtaining a sample free of interference, such as the use of drugs or treatments, because these patients were not deriving physician referral and the majority were without self-medication of vaginal creams.

In our point of view, this would be then the "timing" and the ideal population for us to study the infections called "mutagenic" by Zur Hausen³, singularity to the work.

Observing the data obtained in this population shows low positivity to HPV if compared to the global average, and the high positive rate to *Chlamydia trachomatis* is given by the fact that the average age of the population studied is 25 years old, the age where the highest rate of infection by *Chlamydia trachomatis*, *Herpes simplex* and HPV are, according to Center for Disease Control and Prevention¹².

The work carried out by Murta¹¹ and colleagues in Belo Horizonte-MG found that the figures found to *Chlamydia trachomatis* are concordant to the high positivity rate found in this study.

Despite what was reported in the literature, we did not find significant association between the presence of HPV and vaginal bacterial infections, fungal and parasitic.

Conclusion

Fisher's Chi-square test was applied to socioeconomic data and infections data, but no significant associations were uncovered between the presence of HPV and any of the factors listed above.

Acknowledgments

This manuscript in memory to Silvia Brenna, because this research was his last participation before his death.

Recommendations

Several types of cancer are associated with HPV, joining with parasitic infections occur more injuries. However there is no way to know what people will continue to develop cancer or other health problems. Is important to visit a doctor regularly for checkuping.

References

1. PARKIN D M, BRAY F, FERLAY J, PISANI P. Global cancer statistics, 2002. *CA Cancer J Clin.* 2005;55(2):74-108.
2. Brazilian National Cancer Institute - INCA: 2010 estimative: incidence of cancer in Brazil [Internet]. Rio de Janeiro: Brazilian National Cancer Institute - INCA. 2009. (Results and

discussions summary. cervical cancer; 32-4.). Available at: www1.inca.gov.br/estimativa/2010/estimativa20091201.pdf

3. Zur Hausen H. Papillomaviruses and cancer: from basic studies to clinical application. *Nat Rev Cancer.* 2002; 2(5):342-50.
4. Focaccia R. Veronesi: tratado de infectología. 3 ed. São Paulo: Atheneu; 2005.
5. OHLEMEYER C L, HORNBERGER L L, LUNCH D A, SWIERKOSZ E M . Diagnosis of *Trichomonas vaginalis* in adolescent females: InPouch TV culture versus wet-mount microscopy. *J Adolesc Health.* 1998; Mar;22(3):205-8.
6. JITPRAPHAI P, WISE G J. Pathogenecity of *Trichomonas vaginalis* in the genito urinary tract of experimental male animals. *J Med Assoc Thai.* 1971; Oct;54(10):714-26.
7. Brazilian Health Surveillance Agency. Manual de microbiologia clínica para o controle de infecção em serviços de saúde (Clinical Microbiology Manual for the control of health care infections): preliminary version [Internet]. Brasília: Brazilian Health Surveillance Agency; 2004 [updated in 2004; cited 2011]. Available at: http://bvms.saude.gov.br/bvs/publicacoes/manual_microbiologia_completo.pdf
8. Neto AA, Hamdan JS; Souza RC. Prevalência de cândida na flora vaginal de mulheres atendidas num serviço de planejamento familiar/ Prevalence of candida in the vagina of women attended at a family planning service. *Rev. bras. ginecol. Obstet.* 1999;21(8): 441-445.
9. Hinrichsen SL, Falcão E, Vilella TA, Rêgo L, Lira C, Almeida L, et al. Isolados de Candida em hospital terciário no nordeste do Brasil (Isolated of Candida in a Brazilian Northeast tertiary hospital). *Braz J Microbiol.* 2009;40(2):325-8.
10. Mahony JB, Luinstra KE, Sellors JW, Chernesky MA. Comparison of plasmid- and chromosome-based polymerase chain reaction assays for detecting *Chlamydia trachomatis* nucleic acids. *J Clin Microbiol.* 1993 Jul;31(7):1753-8
11. Murta EF, Souza MA, Araujo Junior E, Adad SJ. Incidence of *Gardnerella Vaginalis*, *Candida* sp and human papilloma virus in cytological smears. *Sao Paulo Med J.* 2000 Jul 6;118(4):105-8.
12. Centers For Disease Control and Prevention. National Center for HIVAIDS, Viral Hepatitis, STD and TB Preventions. Division of STD Preventions <http://www.cdc.gov/std/herpes/>