

The effect of vaginal douching on Pap smear

Vajinal duşun Pap-smear üzerine etkisi

Berfin Can Gök, Soner Gök, Selda Demircan Sezer, Ayhan Atıgan

Gönderilme tarihi:23.03.2022

Kabul tarihi:14.06.2022

Abstract

Purpose: Vaginal douching damages vaginal flora and ecosystem. The disruption of the equilibrium of vaginal flora composition leads to many outcomes having a negative effect on women and newborn health.

Materials and methods: 105 patients who applied vaginal douche were included in this study. All women taken of Pap smear test and the patients were informed in writing about the reasons why they must leave their vaginal douching habits, and a second Pap smear appointment day not earlier than 30 days were given. After taken of patients first Pap smears, none of them took an infection treatment. Pap smear results taken before and after vaginal douche have been compared.

Results: It was seen in the detailed comparison of Pap smear outcomes that the rate of patients with violent inflammation decreased from 11.4% to 1.9%. The decrease in violent inflammation was found statistically significant ($p=0.006$). When the patients with infection were compared regardless of any discrimination of slightly or violent; the rate of the cases with typical infection findings was seen to decrease from 48.5% to 31.4% and found statistically significant ($p=0.01$).

Conclusions: Vaginal flora is a very complex and dynamic component and is under the influence of many factors. Vaginal douching damages vaginal flora structure and balance, and adversely affects women's health in a wide range from a simple vaginal infection to cervical neoplasia.

Key words: Pap smear, vaginal douche, vaginal flora.

Can Gok B, Gok S, Demircan Sezer S, Atıgan A. The effect of vaginal douching on Pap smear. Pam Med J 2022;15:603-609.

Öz

Amaç: Vajinal duş, vajinal floraya ve ekosisteme zarar verir. Vajinal flora dengesinin bozulması, kadın ve yenidoğan sağlığı üzerinde olumsuz etki yapan birçok nedene yol açmaktadır.

Gereç ve yöntem: Çalışmaya vajinal duş uygulayan 105 hasta dahil edildi. Tüm kadınlardan Pap smear testi alındı. Hastalara neden vajinal duş alışkanlıklarını bırakmaları gerektiğine dair yazılı bilgi verildi. 30 günden önce olmamak üzere ikinci bir Pap smear testi için randevu günü verildi. Hastaların ilk Pap smearleri alındıktan sonra hiçbirine enfeksiyon tedavisi verilmedi. Vajinal duştan önce ve sonra alınan pap smear sonuçları karşılaştırıldı.

Bulgular: Pap smear sonuçlarının detaylı karşılaştırmasında şiddetli inflamasyonu olan hasta oranının %11,4'ten %1,9'a düştüğü görüldü. Şiddetli inflamasyondaki azalma istatistiksel olarak anlamlı bulundu ($p=0,006$). Hafif veya şiddetli ayrımına bakılmaksızın enfeksiyonlu hastalar karşılaştırıldığında; tipik enfeksiyon bulgusu olan olguların oranının %48,5'ten %31,4'e düştüğü görüldü ve istatistiksel olarak anlamlı bulundu ($p=0,01$).

Sonuçlar: Vajinal flora; birçok faktörün etkisi altında olan, çok karmaşık ve dinamik bir bileşendir. Vajinal duş, vajinal flora yapısına ve dengesine zarar verir. Basit bir vajinal enfeksiyondan servikal neoplaziye kadar geniş bir yelpazede kadın sağlığını olumsuz etkiler.

Anahtar kelimeler: Pap smear, vajinal duş, vajinal flora.

Can Gök B, Gök S, Demircan Sezer S, Atıgan A. Vajinal duşun Pap-smear üzerine etkisi. Pam Tıp Derg 2022;15:603-609.

Berfin Can Gök, M.D. Denizli Public Hospital, Gynecology and Obstetrics Department, Denizli, Turkey, e-mail: berfinyurdam@gmail.com (<https://orcid.org/0000-0001-5739-3683>)

Soner Gök, Ass. Prof. Pamukkale University, Gynecology and Obstetrics Department, Denizli, Turkey, e-mail: sonerrgok@hotmail.com (<https://orcid.org/0000-0001-8940-1879>)

Selda Demircan Sezer, Prof. Adnan Menderes University, Gynecology and Obstetrics Department, Aydın, Turkey, e-mail: sdemircansezer@gmail.com (<https://orcid.org/0000-0002-2744-5363>)

Ayhan Atıgan, Ass. Prof. Karabuk University, Gynecology and Obstetrics Department, Karabuk, Turkey, e-mail: dratigan@hotmail.com (<https://orcid.org/0000-0002-7257-0593>) (Corresponding Author)

Introduction

Vaginal douching is a flushing process of the vagina with water and similar liquids, for different purposes such as hygiene, religious necessities, and contraception [1, 2]. Vaginal douching is an application, which dates back to old times, widely and traditionally used by women all over the world, and it has a usage rate of 25% at developed countries such as the USA and Canada, and has more than usage rate of 90% at underdeveloped countries such as Zambia and Ghana [3, 4]. As well as its prevalence changes on a regional basis, it is affected by some socio-economic factors such as race and educational level. This application has been seen to be used more widely by black women and people with a low educational level [5]. Although many products are used for vaginal douching, the most widely used product by women is water [6, 7].

Vaginal flora, having an important effect on women and newborn, contains various microorganism communities [8]. Many factors such as age, menstruation, hormonal fluctuation, sexual behaviors, and antibiotics are effective in the composition of vaginal flora [9-12]. The disruption of the balance of vaginal flora composition causes bacterial vaginosis [12, 13]. The disruption of the balance of vaginal flora composition leads to many outcomes having a negative effect on health [14]. Some of those are abnormal obstetric outcomes such as stillbirth, preterm birth, chorioamnionitis, ectopic pregnancy, infertility [15-18]; and development of sexually transmitted infections such as gonorrhea, chlamydia, HPV (human papillomavirus), herpes simplex type 2 and HIV (human immunodeficiency virus) infection [16, 19, 20].

In spite of the fact that there are so many negative side effects of vaginal douching on women's health, it is still widely used especially at communities with low level of education around the world, depending on the factors such as receiving sexual pleasure, wish of tightening vaginal muscles, individualist perception, beliefs, religious [5, 7, 21].

We consider that vaginal douching damages flora and ecosystem thereby increasing vaginal risk and affects Pap smear outcomes adversely. That's why, in this study, we aimed to compare

the Pap smear outcomes of women applying regular vaginal douching, with those of the same women after they have left to apply vaginal douching and to investigate the effects of vaginal douching application on Pap smear outcomes.

Materials and methods

Study design and population

105 patients in the age range between 19-64 years, applying vaginal douching, applied to the gynecology clinic of our hospital, were included in the study. Pregnant, those previously became hysterectomy for any reason, and those received treatment because of genital infection they had within last month were excluded from the study. Each patient was asked whether she was applying vaginal douching, and if she responded yes, then her name, surname, file number, phone number, age, educational level, sexual protection method, fertility status, whether she had a genital infection within last month, if she had, whether she received a treatment or not, frequency and duration of applying vaginal douching, and whether she was using an additional cleanser during vaginal douching were asked and the answers were noted.

In our hospital, the Pap smears of patients are taken of in the term when they have not menstrual bleeding, by previously being informed about not having a vaginal douching, and not using a vaginal drug within the last 72 hours. After an additional questionnaire, medical examination and survey was performed about the patients' complaints and primary problems, the harms of vaginal douching were told, and the patients were informed in writing about the reasons why they must leave their vaginal douching habits, and a second Pap smear appointment day, not earlier than 30 days, were given. After taken of patients' first Pap smears, none of them took an infection treatment. Pap smear test was reported based on the Bethesda system.

This study was approved by the the Adnan Menderes University Clinical Research approved by the Ethics Committee. Before the start of the study, the consent of all individuals was obtained for participation in the study and it was assured that their information would remain confidential.

Data analysis

'SPSS 14.0 for Windows' program' was used for the calculation of statistical analyses. The values were indicated in terms of the average \pm standard deviation (SD) and frequency (%). The McNemar chi-square test was used in the comparison of the values of the two groups. The statistical significance level was regarded as $p < 0.05$.

Results

The average age of 105 patients examined in our clinic was 42.18 ± 10.8 (19-64 years). A great majority of the patients were women in the age range of 40-49 years. Only one woman's age among the women included in the study was below 20 years. The distribution of the patients according to the age groups is given in Table 1.

92.4% case (97 patients) were married, 85.8% (90 patients) multiparous and 14.3% (15 patients) nulliparous. When examined from the educational aspect, more than half of the cases (51.4%) were primary education graduates.

92.4% (97 patients) of the women included in the study stated that they had partners, and were sexually active. The demographical features of the cases are given in Table 2.

The sexual protection methods of the cases were classified as those not using any sexual protection method, coitus interruptus, intrauterine device, preservative, oral contraceptive, bilateral tube ligation and vaginal douching in Table 3. It was observed in our study that, none of the patients used vaginal douching as a sexual protection method.

A great majority of the patients (44.8%) stated that they were using vaginal douching for cleaning at every bath and ritual ablution of the whole body; 31.4% for removing sperm and pre-ejaculatory fluid after having sex; 14.3% for cleansing after relieving oneself, and 9.5% for removing postmenstrual menstrual bleeding. 79% of the cases (83 patients) stated that they were using vaginal douching for five years and longer, 21% (22 patients) for not longer than five years (Table 4).

Table 1. The distribution of the patients according to the age groups

| Age group, y | Patient | Frequency (%) |
|--------------|---------|---------------|
| <20 | 1 | 0.9 |
| 20-29 | 16 | 15.2 |
| 30-39 | 22 | 20.9 |
| 40-49 | 36 | 34.2 |
| >50 | 30 | 28.5 |
| Total | 105 | 100 |

Table 2. The demographical features of the patients

| | Demographical Features | Patient | Frequency % |
|--------------------------|------------------------|---------|-------------|
| Educational level | Illiterate | 9 | 8.6 |
| | Literate | 10 | 9.5 |
| | Primary school | 54 | 51.4 |
| | High school | 26 | 24.8 |
| | Collage | 6 | 5.7 |
| Sexually activity | Sexually active | 97 | 92.4 |
| | Sexually inactive | 8 | 7.6 |
| Fertility | Multiparous | 90 | 85.8 |
| | Nulliparous | 15 | 14.3 |

Table 3. The sexual protection methods of the patients

| Protection methods | Patient | Frequency (%) |
|-------------------------|------------|---------------|
| No using any method | 41 | 39 |
| Coitus Interruptus | 29 | 27.6 |
| Intrauterine device | 11 | 10.5 |
| Preservative | 10 | 9.5 |
| Oral contraceptive | 10 | 9.5 |
| Bilateral tube ligation | 4 | 3.8 |
| Vaginal douching | - | - |
| Total | 105 | 100 |

Table 4. Reasons and times for patients to have vaginal douche

| Times | Patient | Frequency (%) |
|--------------------------|------------|---------------|
| Every bath | 47 | 44.8 |
| After sexuality | 33 | 31.4 |
| Every toilet | 15 | 14.3 |
| After every menstruation | 10 | 9.5 |
| Total | 105 | 100 |

From the aspect of material used during vaginal douching; 68.6% patients (72 cases) were using only water, 17.1% (18 cases) water and soap, 14.3% (15 cases) water and shower gel. It was seen in the detailed comparison of Pap smear outcomes that the rate of patients with slight inflammation decreased from 39 (37.2%) to 31 (29.5%). It was also seen that the rate of patients with violent inflammation decreased from 12 (11.4%) to 2 (1.9%). While the decrease in slightly inflammation was found not statistically significant ($p>0.05$), that in violent inflammation was found statistically significant ($p=0.006$). When the patients with infection were compared regardless of any

discrimination of slightly or violent; the rate of the cases with typical infection findings was seen to decrease from 51 (48.5%) to 33 (31.4%), and found statistically significant ($p=0.01$) (Table 5). On the other hand, the first outcomes of Pap smear show that the quantity of taken of Pap smear of the patients with atrophy and ASCUS decreased after they have left vaginal douching application, but the decrease was found not significant ($p>0.05$). While the number of a patient diagnosed with LSIL was one, it rose up two after leaving vaginal douching application, but this difference was not significant ($p>0.05$) (Table 5).

Table 5. Detailed analysis of the Pap-smear outcomes of the patients, before they got started the vaginal douching application and after they have left vaginal douching application

| Pap-smear outcomes | Vaginal douching n=105 (%) | Left Vaginal douching n=105 (%) | p^* |
|-----------------------|-------------------------------|------------------------------------|--------------|
| Normal | 35 (33.3) | 57 (54.2) | 0.002 |
| Slightly inflammation | 39 (37.2) | 31 (29.5) | 0.186 |
| Violent inflammation | 12 (11.4) | 2 (1.9) | 0.006 |
| Infection | 51 (48.5) | 33 (31.4) | 0.01 |
| ASCUS | 10 (9.5) | 6 (5.7) | 0.29 |
| Atrophy | 7 (6.6) | 6 (5.7) | 0.77 |
| AGUS | 1 (0.9) | 1 (0.9) | 1 |
| LSIL | 1 (0.9) | 2 (19.0) | 0.56 |

* $p<0.05$ was regarded as significant, McNemar chi-square test

Discussion

In this study, the Pap smear outcomes of women applying regular vaginal douching were compared with those of the same women after they have left to apply vaginal douching, and the decrease in infection findings was observed in the same outcomes gotten after they have left vaginal douching application.

In our study, 34.2% (36 cases) of the women having a vaginal douche was in the age range of 40–49 years. The great majority of the women were primary education graduates (54 persons (51.4%). The relevant studies show that age, education, ethnic origin, and socioeconomic status affect the frequency of having vaginal douche. The frequency of having vaginal douche has been seen to increase significantly in the group having low socioeconomic status and low education [5]. It was found that vaginal prevalence is higher at groups with low education, among both Afro American and white women. In our study, 33 (31.4%) of the women included in our study stated to have vaginal douche after sexual intercourse, 10 women (9.5%) at the end of menstrual bleeding, 47 women (44.8%) at every bath, 15 women (14.3%) every time when they make their toilet. A relevant study shows that 60% of the women having vaginal douche stated to have vaginal douche to feel clean and hygienic themselves [22]. Another relevant study base on the reasons for women's having vaginal douche on two conceptual models. These are cosmetic model and infection control model [23]. In the cosmetic model, women have vaginal douche to feel clean themselves after menstrual bleeding and to make their partners happy. In the infection control model, it is considered to move the bacteria away that will likely to cause infection and irritation during vaginal douching, thus, to play a significant role in the treatment of infections and provide protection against infections. 72 (68.6%) of the women included in our study stated to use only water during vaginal douching, 18 (17.1%) women water and soap, 15 women shower gel. The relevant studies show that the most widely used product for vaginal douching is water [7, 22, 24]. In addition to water, a great variety of materials existing from the thought of making oneself feel good, ranging from commercial materials to natural and herbal ones, are used. American women mostly prefer commercial products, African women herbal products [25].

Our study findings show that vaginal douching application affects the outcomes of a Pap smear. This change has been considered to arise depending on the change in vaginal flora in women having vaginal douche. It was found that the infection rate was seen in 51 women (48.5%) in the outcomes of Pap smear gotten at the vaginal douching period was seen to decrease to 33 (31.4%), in the outcomes of Pap smear gotten after these women have left the vaginal douching application. Recent technological progress has shown that the vagina has a dynamic and complex ecosystem, and its flora prevalence is created by *Lactobacillus* species [12]. These *Lactobacillus* species generate acidic pH in the vagina through lactic acid and hydrogen peroxide thereby creating an ecosystem protecting against harmful microbes [26]. Vagina flora, under the dominance of *Lactobacillus*, is affected by factors such as age, menstruation, pregnancy, vaginal douching [27]. In situations like vaginal douching, when the balance of flora is disrupted, *Lactobacillus* species that protect the vaginal ecosystem decrease, pathogen microorganisms such as *Gardnerella vaginalis*, *Bacteroides* spp., *Mobiluncus* spp. and anaerobic cocci increase, and accordingly, bacterial vaginosis, a widely seen gynecologic disease, occurs [26, 28]. Bacterial vaginosis has many side effects adversely affecting women's health [14]. Due to bacterial vaginosis; an increase in having an abortion, preterm delivery, chorioamnionitis, pelvic inflammatory disease risk can be seen [15]. A relevant study done shows that vaginal douching increases high-risk HPV infection, and as a consequence, it causes HPV-related genital cancer risk to increase [16, 29, 30]. Another relevant study shows that a decrease in normal vaginal commensal microbiota increases the damage risk in the vaginal epithelium that excretes IL-33, which is a cytokine that prevents effector T cells from migrating to vaginal mucosal tissue, thereby blocking the production of interferon-gamma, a major cytokine in the mucosal antiviral defense [31]. As a result, vaginosis-associated local antiviral defense is pressed, and an increase in invasive pathogens like HIV is observed [32].

In conclusion, vaginal flora is a very complex and dynamic component and is under the influence of many factors. Vaginal douching damages vaginal flora structure and balance,

and adversely affects women's health in a wide range from a simple vaginal infection to cervical neoplasia.

In consequence, in the light of relevant studies, vaginal douching is regarded as a traditional application having harmful effects on women's health and is not supported. Especially, the habit of vaginal douching should be investigated in the scope of primary healthcare by monitoring women, pregnant and puerperal in the age group of 15-49 years, and vaginal douche applicants should be trained. But, changing habits like vaginal douching, which is affected by many and traditional factors, is not as easy as it is supposed. Considering individual and sociological interactions of health-related behaviors, and decisiveness of religious effect, this habit can be preventable through multidisciplinary and intersectoral collaboration, and society based, special education programs aiming to form behavioral change.

Conflict of interest: No conflict of interest was declared by the authors.

References

- Sevil S, Kaplan S, Ünsal A, Abay H, Pınar G, Nazlı Y. Vaginal douching among married Turkish women and relation to quality of life. *Indian J Tradit Know* 2016;15:611-618. Available at: <http://nopr.niscair.res.in/handle/123456789/35240>. Accessed March 24, 2022
- Martino JL, Youngpairoj S, Vermund SH. Vaginal douching: personal practices and public policies. *J Womens Health* 2004;13:1048-1065. <https://doi.org/10.1089/jwh.2004.13.1048>
- Alcaide ML, Chisembele M, Mumbi M, Malupande E, Jones D. Examining targets for HIV prevention: intravaginal practices in Urban Lusaka, Zambia. *Aids Patient Care St* 2014;28:121-127. <https://doi.org/10.1089/apc.2013.0309>
- Ekpenyong CE, Daniel NE, Akpan EE. Vaginal douching behavior among young adult women and the perceived adverse health effects. *J Public Health Epidemiol* 2014;6:182-191. <https://doi.org/10.5897/JPHE2014.0622>
- Annang L, Grimley DM, Hook EW. Vaginal douche practices among black women at risk: exploring douching prevalence, reasons for douching, and sexually transmitted disease infection. *Sex Transm Dis* 2006;33:215-219. Available at: <http://www.jstor.org/stable/44971186>. Accessed March 24, 2022
- Branch F, Woodruff TJ, Mitro SD, Zota AR. Vaginal douching and racial/ethnic disparities in phthalates exposures among reproductive-aged women: national health and nutrition examination survey 2001-2004. *Environmental Health* 2015;14:1-8. <https://doi.org/10.1186/s12940-015-0043-6>
- McCarthy FNA, Nii Trebi NI, Musah BO, Asmah RH. Intravaginal practices and lactobacilli colonization among women in Accra, Ghana. *BMC Women's Health* 2015;15:46(e1-5). <https://doi.org/10.1186/s12905-015-0205-2>
- Li JR, McCormick J, Bocking A, Reid G. Importance of vaginal microbes in reproductive health. *Reprod Sci* 2012;19:235-242. <https://doi.org/10.1177/1933719111418379>
- Zapata HJ, Quagliarello VJ. The microbiota and microbiome in aging: potential implications in health and age-related diseases. *J Am Geriatr Soc* 2015;63:776-781. <https://doi.org/10.1111/jgs.13310>
- Herbst Kralovetz MM, Pyles RB, Ratner AJ, Sycuro LK, Mitchell C. New systems for studying intercellular interactions in bacterial vaginosis. *J Infect Dis* 2016;214:6-13. <https://doi.org/10.1093/infdis/jiw130>
- Fettweis JM, Brooks JP, Serrano MG, et al. Differences in vaginal microbiome in African American women versus women of European ancestry. *Microbiology* 2014;160:2272-2282. <https://doi.org/10.1099/mic.0.081034-0>
- Mendling W. Vaginal microbiota. *Adv Exp Med Biol* 2016;902:83-93. https://doi.org/10.1007/978-3-319-31248-4_6
- Reid G. Is bacterial vaginosis a disease? *Appl Microbiol Biot* 2018;102:553-558. <https://doi.org/10.1007/s00253-017-8659-9>
- Martin DH, Marrazzo JM. The vaginal microbiome: current understanding and future directions. *J Infect Dis* 2016;214:36-41. <https://doi.org/10.1093/infdis/jiw184>
- Afolabi BB, Moses OE, Oduyebo OO. Bacterial vaginosis and pregnancy outcome in Lagos, Nigeria. *Open Forum Infectious Diseases* 2016;3:ofw030. <https://doi.org/10.1093/ofid/ofw030>
- Bui TC, Thai TN, Tran LTH, Shete SS, Ramondetta LM, Basen Engquist KM. Association between vaginal douching and genital human papillomavirus infection among women in the United States. *J Infect Dis* 2016;214:1370-1375. <https://doi.org/10.1093/infdis/jiw388>
- Bui TC, Tran LTH, Ross MW, Markham CM. Douching practices among female sex workers in Phnom Penh, Cambodia. *Int J Std Aids* 2015;26:238-242. <https://doi.org/10.1177/0956462414533098>

18. Attieh E, Maalouf S, Roumieh D, Abdayem P, AbiTayeh G, Kesrouani A. Feminine hygiene practices among female patients and nurses in Lebanon. *Reprod Health* 2016;13:59. <https://doi.org/10.1186/s12978-016-0182-4>
19. Srinivasan S, Fredricks DN. The human vaginal bacterial biota and bacterial vaginosis. *Interdiscip Perspect Infect Dis* 2008;2008:750479. <https://doi.org/10.1155/2008/750479>
20. McClelland RS, Lingappa JR, Srinivasan S, et al. Evaluation of the association between the concentrations of key vaginal bacteria and the increased risk of HIV acquisition in African women from five cohorts: a nested case-control study. *Lancet Infect Dis* 2018;18:554-564. [https://doi.org/10.1016/S1473-3099\(18\)30058-6](https://doi.org/10.1016/S1473-3099(18)30058-6)
21. Yaman HE, Aygun M, Tosun H. Traditional genital hygiene practices in Turkey. *Indian J Tradit Know* 2016;15:214-218. Available at: <http://hdl.handle.net/123456789/33969>. Accessed 24.03.2022
22. Ziba FA, Yakong VN, Asore RA, Frederickson K, Flynn M. Douching practices among women in the Bolgatanga municipality of the upper east region of Ghana. *BMC Women's Health* 2019;19:32(e1-5). <https://doi.org/10.1186/s12905-019-0720-7>
23. Mckee MD, Baquero M, Anderson M, Alvarez A, Karasz A. Vaginal douching among Latinas: practices and meaning. *Matern Child Hlth J* 2009;13:98-106. <https://doi.org/10.1007/s10995-008-0327-3>
24. Brown JM, Poirot E, Hess KL, Brown S, Vertucci M, Hezareh M. Motivations for intravaginal product use among a cohort of women in Los Angeles. *Plos One* 2016;11:e0151378. <https://doi.org/10.1371/journal.pone.0151378>
25. Brotman RM, Klebanoff MA, Nansel TR, et al. A longitudinal study of vaginal douching and bacterial vaginosis - a marginal structural modeling analysis. *Am J Epidemiol* 2008;168:188-196. <https://doi.org/10.1093/aje/kwn103>
26. Africa CWJ, Nel J, Stemmet M. Anaerobes and bacterial vaginosis in pregnancy: virulence factors contributing to vaginal colonisation. *Int J Env Res Pub He* 2014;11:6979-7000. <https://doi.org/10.3390/ijerph110706979>
27. Bautista CT, Wurapa E, Sateren WB, Morris S, Hollingsworth B, Sanchez JL. Bacterial vaginosis: a synthesis of the literature on etiology, prevalence, risk factors, and relationship with chlamydia and gonorrhea infections. *Military Med Res* 2016;3:4(e1-10). <https://doi.org/10.1186/s40779-016-0074-5>
28. Muzny CA, Schwebke JR. Pathogenesis of bacterial vaginosis: discussion of current hypotheses. *J Infect Dis* 2016;214:1-5. <https://doi.org/10.1093/infdis/jiw121>
29. Moscicki AB, Ma YF, Farhat S, et al. Redetection of cervical human papillomavirus type 16 (HPV16) in women with a history of HPV16. *J Infect Dis* 2013;208:403-412. <https://doi.org/10.1093/infdis/jit175>
30. Dickson EL, Vogel RI, Bliss RL, Downs LS. Multiple-type human papillomavirus (HPV) infections a cross-sectional analysis of the prevalence of specific types in 309,000 women referred for HPV testing at the time of cervical cytology. *Int J Gynecol Cancer* 2013;23:1295-1302. <https://doi.org/10.1097/IGC.0b013e31829e9fb4>
31. Oh JE, Kim BC, Chang DH, et al. Dysbiosis-induced IL-33 contributes to impaired antiviral immunity in the genital mucosa. *P Natl Acad Sci USA* 2016;113:762-771. <https://doi.org/10.1073/pnas.1518589113>
32. Bayigga L, Kateete DP, Anderson DJ, Sekikubo M, Nakanjako D. Diversity of vaginal microbiota in sub-Saharan Africa and its effects on HIV transmission and prevention. *Am J Obstet Gynecol* 2019;220:155-166. <https://doi.org/10.1016/j.ajog.2018.10.014>

Acknowledgments: The authors thank all the participants who helped us in the collection of samples.

Ethics committee approval: Adnan Menderes University Clinical Research approved by the Ethics Committee (date: 30.06.2010 and decision no: 3).

Contributions of the authors to the article

B.C.G., S.D.S. set up the main idea and hypothesis of the study. B.C.G., S.D.S. and S.G. developed the theory and edited the material method section. A.A. made the evaluation of data in results section. The discussion part of the article was written by B.C.G., S.G. and A.A. reviewed, made necessary corrections and approved. In addition, all authors discussed the entire study and approved its final version.