

Article

Vulvovaginal Hygiene Practices Among Pregnant Ladies in Rural West Bengal: A Cross sectional Observational Study

Partha Sarathi Mitra¹, Nupur Ghosh², Avik De³, Monalisa Chatterjee⁴, Pamela Khatua⁴, Khwaja Alim⁵

¹Department of Obstetrics and Gynaecology, Kolkata medical College and Hospital, Kolkata

²Department of Obstetrics and Gynaecology, Burdwan medical College and Hospital, Bardhaman

³WBMES, Burdwan Medical College and Hospital, Burdwan.

⁴Dept. of Gynecology & Obstetrics, Burdwan Medical College, Burdwan

⁵WBHS, W. B.

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CORRESPONDENCE

Phone: 7980115887

E-mail: parthasarathimitra1972@gmail.com

A B S T R A C T

Vulvovaginitis is very common in pregnancy in our country. Lactobacilli are the normal vaginal microflora which maintains acidic vaginal pH. Lack of hygiene may lead to colonisation of coliforms resulting in vulvovaginitis which in turn may cause serious health problems for the mother and the new-born. A cross-sectional observational study was conducted on pregnant women attending antenatal OPD of BMCH on every Wednesday between 1st July to 31st December, 2022 with a preformed questionnaire which dealt with different parameters to observe whether present pregnancy was complicated by any vulvovaginal ailments. Although they had little knowledge about vulvovaginal microbiota, pathogenesis and morbidity of vulvovaginitis during pregnancy, most had a practice of vulvovaginal cleaning with plain water and not commercial douching. Protection against vulvovaginal infection is particularly important for pregnant women as it increases the risk serious maternal and neonatal morbidities. Routine washing of the vulva with plain water is beneficial to maintain cleanliness and prevent vulvovaginal infection. Vulvar cleansing products does not treat infections rather disrupts normal vaginal microbiota thus promotes colonization of uropathogens. Prevention of vulvovaginal infection by simple hygiene maintenance can be achieved by educating antenatal mothers during their visits.

I. INTRODUCTION

Vulvovaginal disease is often caused by multiple factors, such as immune deficiency, hormonal changes, stress, or use of a vaginal douche or soap to clean the vagina which may upset the normal flora and cause infections. The vagina has no gland. It is lubricated by transudates from subepithelial capillary plexus across the epithelium which is permeable. The pH of vagina at reproductive age is 3.8–4.4 (F. Gary, 2018). The female genital tracts harbour a wide variety of microflora² which are commensals and mostly constituted by lactobacilli³, without causing infection⁴. However, they may become pathogenic in case of disorder in the normal microflora⁵. Vulval microflora are lipophilic and non-lipophilic diphtheroids; coagulase negative staphylococci, micrococci, lactobacilli; streptococci; gram-negative rods; gram-negative bacilli; neisseria; gardnerella vaginalis; and/or yeasts. Vaginal microflora consists of lactobacillus spp., atobium vaginae, megasphaera spp., leptotrichia spp., gardnella vaginalis, staphylococcus aureus, and/or candida albicans (F. Gary, 2018), the vulvovaginal vasculature develops varicosities during pregnancy from increased venous pressure by enlarged uterus. Due to increased vascularity vaginal secretions are notably increased. The pH becomes acidic due to more conversion of glycogen into lactic acid by lactobacillus acidophilus consequent to high estrogen level of pregnancy and subsequently this acidic pH prevents multiplication of pathogenic organisms. Vulvovaginitis and urinary tract infection are very common in pregnancy in our country, occurs when there is a shift in the normal flora dominated by lactobacilli to coliform uropathogens (Yazici S, 2009; Olsen BE et al, 2000; Mignini L et al, 2009), and can result in serious health problems for the mother and the new-born (Mignini L et al, 2009; Pete PMN et al 2019) such as pyelonephritis, early and mid-trimester pregnancy loss, low birth weight, preterm labor, preterm premature rupture of membrane, intra uterine growth restriction, and increased incidence of perinatal death (Schnarr J et al, 2008; Al-Badr A et al, 2013; Hillier SL et al, 1995; Fiscella K et al 1998).

II. METHODS

A cross-sectional observational study was conducted on pregnant women attending antenatal out patients' department (OPD) of Burdwan Medical College and Hospital on every Wednesday between 1st July and 31st December, 2022. which is a tertiary care hospital located in Bardhaman, a suburban town in the district of Purba Bardhaman, West Bengal, where patients from both urban and rural areas regularly attend. Average number of patients attending OPD during our study on each Wednesdays of every week was 110 to 140. Every fifth antenatal mothers of at least six weeks of pregnancy were selected, counseled and requested for an interview. Those who volunteered were enrolled. Total number of 482 (out of 780 approached) patients were recruited. A clear and thorough explanation of the objectives and nature of the study was provided and the participant was invited to take part in the study. A semi-structured self-prepared preformed questionnaire was explained in their own language, which dealt with different parameters such as socio-demographic characteristics, awareness of importance of vulvo-vaginal-perineal hygiene and usual practices of vulvo-vaginal-perineal washing, medicated or indigenous douching, the use of antiseptic solution, dressing and underwear used, sexual habits and practices in antenatal period and hygiene practices after sexual act. History was taken about her past (if any) and present pregnancy like parity, previous pregnancy loss and duration of present pregnancy, whether the present pregnancy was complicated by any vulvovaginal or lower genitourinary ailments. The data collected was tabulated in prescribed format. Statistical analysis was done using microsoft excel chi-test.

III. RESULT

In this study we have recruited 482 participants, most of them were teen age pregnancies, 361 out of 482, ie. 74.89% patients in 15 – 25 yrs age group, although 8.29% was in elderly

mothers' group also. Most of them have completed their primary school education i.e. 67.8%, a few are illiterate and 21.16% have completed their high school. As our hospital is located in a suburban area, we provide service to a wide range of urban and rural areas. We found 65.9% of our candidates from rural, 20.9% from semi urban and 13% from urban areas including slums. Religion is almost equally distributed between Hindus and Muslims among the attendees. Excepting a few exceptions more than 97% were married, a bit over 50% were housewives, while the rest were economically independent by either service or daily labour activities. About 62.65% belong to lower socio-economic condition and a few in the upper or middle class society. The modern era revolution has brought the privilege of household sanitary latrine to more than 90% of our study population. 95.64% of our study subjects had no addiction, 3.94% had history of smoking (bidi or chutta) and 5.19% was addicted to alcoholic beverages. (Table-1)

Table 1. Participants' demographic data (n=482)

Age	15 – 25 yrs	361	74.89%
	26 – 35 yrs	81	16.8%
	36 – 45yrs	40	8.29%
Education	No schooling	53	10.9%
	Lower school	327	67.8%
	Higher school	102	21.16%
Residence	Rural	318	65.9%
	Semi urban	101	20.9%
	Urban	63	13%
Religion	Hindu	272	56.4%
	Muslim	199	41.28%
	Other	11	2.28%
Marital status	Married	470	97.51%
	Cohabitation	8	1.65%
	Single	4	0.82%
Occupation	Housewife	265	54.97%
	Service	39	8.09%
	Labour	178	36.92%

Socio economic status	Upper	49	10.16%
	Middle	131	27.17%
	Lower	302	62.65%
SanitaryLatrine/ Washroom	No	09	1.86%
	Yes	434	90.04%
	Don't use	39	8.09%
Substance abuse	No	461	95.64%
	Smoking	19	3.94%
	Alcohol	25	5.19%

Most of our study population was second gravida mothers and the majority was in their third trimester pregnancy. 40.66% had no history of spontaneous abortion, 40.87% had one and 18.46% had two or more spontaneous abortion in their past obstetric history (Table-2).

Table 2. Participants’ obstetric history and clinical symptoms (n=482)

Parity	Para 0	120	24.89%
	Para 1 to 3	297	61.61%
	Para >4	65	13.48%
Duration of pregnancy	6 to 12 wks	32	6.63%
	12 to 24 wks	119	24.68%
	24 to 36 wks	331	68.76%
Previous history of SA	No SA	196	40.66%
	One SA	197	40.87%
	SA >2	89	18.46%

Many of them gave history of vulvovaginal symptoms i.e. vaginal discharge (10.99%), pruritus vulvae (7.05%), burning sensation (18.87%) and rest asymptomatic (63.07%). Statistical test of significance for the clinical vulvo vaginal and urinary symptoms among the participants was done using microsoft excel chi-test software. The chi square p value of these clinico pathological symptoms is too high and we will not reject the null hypothesis (Chi square test p-value 1.8001E-83). As far as urinary symptoms are concerned there were no symptoms in 87.73%, asymptomatic

bacteriuria in 6.43% and symptomatic bacteriuria in 6.01% patients in the study group (Chi square test p-value 4.389E-195) (Table-3)

Table 3. Participants’ clinical symptoms (n=482)

Vulvo vaginal symptoms	No Symptoms	304	63.07%	Chi square test p-value 1.8001E-83
	Vaginal discharge	53	10.99%	
	Pruritus vulvae	34	7.05%	
	Vulvovaginal burning sensation	91	18.87%	
Urinary symptoms	Asymptomatic, no bacteriuria	422	87.73%	Chi square test p-value 4.389E-195
	Asymptomatic bacteriuria	31	06.43%	
	Symptomatic bacteriuria	29	06.01%	

As far as vulvovaginal and urinary symptoms are concerned, in both cases **Chi square test p value is 4.389(>.05)**. Chitest p-value is too high so that we are not going to reject the null hypothesis ie the observed symptoms in our study population is not statistically significant and we have found it merely by chance (p value for the chi square test less than or equal to .05 was regarded as evidence of a statistically significant result).

On questioning about their habits, we found most of them i.e.57-58% use simple water for washing vulvovaginal area instead of medicated douching, which was only 0.82% Majority of the pregnant mothers avoided wearing undergarments while only 1.64% had habit of unhygienic underwear use. 80.70% practiced household bathing in shower regularly while 12.03% in common water bodies. We also asked about pattern of sexual activity and practice after sexual exposure and found majority preferred infrequent and protected intercourse during pregnancy (72.40%). Here also 62.28% preferred only water for vulvovaginal wash after the act of physical contact (Table-4).

Table 4. Vulvovaginal hygiene maintenance practices among the study population (n=482)

Practice of hygiene at Vulva and Perineum	Nil	17	3.52%
	Water	278	57.67%
	Soap and water	183	37.96%
	Medicated douching	04	0.82%
Practice of hygiene at Vagina	Nil	17	3.52%
	Water	282	58.5%

	Soap and water	183	37.96%
	Medicated douching	00	00%
Use of undergarments	No undergarments	360	74.68%
	Tight undergarments	23	4.77%
	Loose undergarments	91	18.87%
	Unhygienic undergarments	08	1.65%
Bathing practices	Avoids bathing	06	1.24%
	Bath at common waterbodies	58	12.03%
	Household bathing in shower	389	80.70%
	Not soaking the whole body	29	6.01%
Sexual practices	Avoids intercourse	79	16.39%
	Frequent unprotected intercourse	37	7.67%
	Infrequent & protected (>once/wk)	349	72.40%
	Frequent & protected (<once/wk)	17	3.52%
Practice of hygiene after sexual activity N = 403	Nil	103	25.55%
	Water	251	62.28%
	Soap and water	45	11.91%
	Medicated douching	04	0.995

In Table-5 we statistically analysed the data to find any correlation between vulvovaginal hygiene maintenance and vulvovaginal-urinary symptoms and found The p value too high (Chi square test p-value 1.41371E-94) to declare no significant correlation between practice of vulvovaginal hygiene and vulvovaginitis. Similarly we found statistically non significant association between vulvovaginal hygienic practice and incidence of UTI (Chi square test p-value 6.61609E-94) as obtained by interview in our study subjects of antenatal mothers.

Table 5. Correlation between vulvovaginal hygiene maintenance and vulvovaginal-urinary symptoms

S/S of Vulvovaginitis N=304	No	S/S of Vulvovaginitis N=178
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Vulvovaginal hygiene maintenance N=461	hygiene practice	284	177	Chi square test p-value 1.41371E-94
No Vulvovaginal hygiene maintenance N=21	hygiene practice	20	01	
		UTI and ASB N=60	Asymtomaic & no UTI N=422	
Vulvovaginal hygiene practice N=461	perineal maintenance	39	422	Chi square test p-value 6.61609E-94
No Vulvovaginal hygiene maintenance N=21	perineal maintenance	18	03	

The p value in both the cases are more than .05 which means we have found no significant correlation between practice of vulvovaginal hygiene as obtained by interview, has got no significant statistical correlation with vulvovaginitis and UTI in our study subjects of antenatal mothers.

IV. DISCUSSION

The vulva is the first line of defense against genital tract infection. Collection of moisture, sweating & menstrual blood in the vulvar folds and hormonal fluctuations influence vulvar microbial growth, potentially resulting in foul smell as well as vulvovaginal infection. Vulvar skin differs from other body parts in respect of hydration, friction, permeability, irritability and is more susceptible to topical agents (Holst E, Goffeng AR, Andersch B, 1994 ; Hay PE et al, 1994 ; Chen Y, Bruning E, Rubino J, Eder SE, 2017). The non-keratinized vulvar vestibule is likely to be more permeable than keratinized skin (Holst E, Goffeng AR, Andersch B, 1994). Genital skin is unique in that it is covered by a thin stratum corneum containing large hair follicles, making it easier for microbial and other substances to permeate the skin (Farage MA and Maibach HI, 2016). The vagina is a fibro muscular canal extending from its external opening in the vulva to the cervix and is composed mainly of smooth muscle covered with a non-keratinized epithelial lining which is kept moist by fluid secreted through the vaginal wall and mucus from cervical & vestibular glands until the menopause. The tight junctions of vaginal mucosa along with thick cervical mucus act as a physiological barrier against microorganisms during pregnancy. Abundance of Lactobacillus in pregnancy reduce vaginal pH and increase vaginal secretions which act as a barrier against pathogenic microbes too (Farage MA, 2005). The major changes in the vaginal microbiome occur during early pregnancy, it gets back to baseline i.e., decrease in Lactobacilli and enrichment of bacterial associates during later stages of pregnancy and puerperium (Wakashin K, 2007). Several studies have reported that there is alteration and physiological changes during pregnancy which decreases the ability of the lower genitourinary tract to resist invading bacteria (Farage MA, 2005). Since long time it has been believed absence of lactobacilli leave vagina susceptible to infections leading to vulvovaginal and lower genitourinary tract infection followed by upper genitourinary tract infection resulting in preterm birth and pregnancy complications.

Along with endocrinological, immunological and metabolic changes during pregnancy, change in environment, weight, diet pattern, hormonal milieu and mostly knowledge, attitude and practices towards vulvovaginal-perineal hygiene during pregnancy can cause significant alterations in the microbiome (Wakashin K, 2007). The hormonal changes during pregnancy, rising progesterone and estrogen levels often lead to numerous physiological effects which may affect the microbiome composition. Microbiota is also influenced by changes in metabolism, as noted in obesity, metabolic syndrome, and diabetes. Thus, the metabolic changes in pregnancy are expected to influence the composition of microbiota. Health and hygiene practice during pregnancy is an important factor of perinatal and long term maternal and child health (Wakashin K, 2007). Our research aimed at highlighting knowledge of genital hygiene including personal hygiene behaviors and practices among pregnant women who were recruited in the study. Apart from antenatal care, diet and therapies, vulvovaginal hygiene are key components that need to be followed up and monitored during pregnancy. Vaginal douching practices may vary according to cultural factors and can be transmitted from one generation to the other. This was observed in the current study where the majority of participants declared that they received information on genital hygiene from a family member. In Africa, black women believed the vagina contains germs, which is the reason for the widespread practice of douching (MacIntyre DA et al, 2015). Other studies reported vaginal douching and use of antiseptic agents as a major factor in the outcome of reproductive and gynecologic health problems (Ramos BA et al, 2015; Hull T, 2011; Cottrell BH, 2010), this study we found most of the antenatal mothers, 57.67% of the total used plain water for washing vulvovaginal area and perineum and 37.96% used non-irritating agents like simple soap-water for vulva cleaning. Antiseptic solutions with water were preferred in only 0.82% for only vulval & perineal area but none used antiseptic for vaginal cleaning, it was found that apart from a few 3.52%, who never cared for genital hygiene, most women perceived their genital region as most susceptible for pathogenic organism growth and paid more attention. But it was also found that indigenous or unformulated antiseptic intra vaginal douching and cleaning in apprehension of vaginal hygiene might be a risky practice for the vaginal microflora as reported by other authors (Klebanoff MA et al, 2010; Fashemi B et al, 2013, Rothman KJ et al, 2003). Vaginal douching had no known confirmed health benefits, might undermine the innate immune defenses by altering the normal vaginal flora and thus predisposed women to infections like pelvic inflammatory diseases (PID), sexually transmitted infection (STI) and also endometriosis (Rosenberg MJ, Phillips RS, Holmes MD, 1991; Rosenberg MJ, Phillips RS, 1992; Sutton MY et al, 2006; Hutchinson KB, Kip KE, Ness RB, 2007; Ness RB et al, 2002; Scholes D et al, 1998).

Scholes and collaborators (2000), in their study on the risk factors for recurrent urinary tract infection in young women cited wearing tight undergarments among factors predisposing women to recurrent UTIs (Scholes D et al, 1993). In this study, 4.77% of participants declared wearing tight undergarments, 74.68% used none and 1.65% are most vulnerable for vulvovaginal urinary infection as they are too reluctant or casual to change their undergarments at frequent interval which indicates lack of good integration and practice of health promotion advice provided to women during antenatal visits (Scholes D et al, 1993).

Taking a bath by soaking the whole body in water could have potential genital adverse effects if the water used is of poor quality, and it was important to know the extent of this practice. Overall, three different bathing habits were found among participants: shower, soaking the whole body in water either in common ponds or tube well or municipal supply. It is well documented

from different studies that pelvic inflammatory disease, vulvovaginitis, UTI, etc. are often associated with bathing in common water bodies, and patients were always counseled to avoid such practices in rural area. Further studies should be conducted to identify risky types of bathing and their course.

Patterns of sexual exposure, habits and practice of hygiene after sexual activity during pregnancy are very important factors and data were collected by interview. Routine washing of the vulva after sexual activity is desirable to prevent accumulation of vaginal discharge, body fluids, sweat, urine, and fecal contamination. Although vulvar cleansing may be a useful adjunct to medical treatment, commercial or indigenous vulvar cleansing products for cleanliness and odor control may upset vulvovaginal pH and vulvovaginal microflora needed for protection against infection (Cottrell BH, 2010). In 2011, the Royal College of Obstetricians and Gynaecologists (RCOG) published evidence-based guidelines for care of vulvar skin disorders (Box 1) (Holst E, Goffeng AR, Andersch B, 1994; Wølner-Hanssen P, 1990).

Box 1. RCOG Guidance on Care of Vulvar Skin

Box 1. RCOG guidance on care of vulvar skin.	
1	Most women with a vulvar disorder (e.g. contact dermatitis, vulvovaginitis) need advice about vulvar skin care and how to avoid contact irritants.
2	Washing with water can cause dry skin and make itching worse. Use a small amount of soap substitute and water to clean the vulva.
3	Shower rather than bathe and clean the vulva only once a day. Overcleaning can aggravate vulvar symptoms (e.g. symptoms of contact dermatitis). An emollient may be helpful.
4	Avoid using sponges or flannels. Just use your hand. Gently pat dry with a soft towel.
5	Wear loose-fitting silk or cotton underwear. Avoid close-fitting clothes. Wear loose-fitting trousers or skirts and replace tights with stockings. You may prefer to wear long skirts without underwear.
6	Sleep without underwear.
7	Avoid fabric conditioners and biological washing powders. Consider washing underwear separately in a non-biological laundry detergent.
8	Avoid using soap, shower gel, scrubs, bubble bath, deodorant, baby wipes, or douches on the vulva.
9	Some over-the-counter creams, including baby or nappy creams, herbal creams (e.g. tea tree oil, aloe vera), and “thrush” treatments, may include irritants.
10	Avoid antiseptic (as a cream or added to bath water) in the vulvar area.
11	Avoid using panty liners or sanitary towels on a regular basis. • Avoid antiseptic (as a cream or added to bath water) in the vulvar area.
12	Wear white or light colored underwear. Dark textile dyes (black, navy) may cause an allergy, but if new underwear is laundered before use, it will be less likely to cause a problem.
13	Avoid using colored toilet paper.
14	Avoid wearing nail varnish on fingernails if you tend to scratch your skin

Middle East and Central Asia (MECA) recommended on female genital hygiene (Box 2) (Holst E, Goffeng AR, Andersch B, 1994; Scholes D, H.M.Thomas, Roberts L. Pacita, Stapleton E Ann, 2000). Both guidelines suggest daily vulva cleansing with a gentle hypoallergenic liquid wash avoiding soap, shower gel, scrubs, bubble bath, deodorant, baby wipe or douches on the vulva and recommend postpartum care should include frequent cleansing, drying, using pads as necessary to maintain dryness over any sutures without any creams.

Box 2. MECA Guidelines on Female Genital Hygiene

Box 2. MECA guidelines on female genital hygiene.	
1	Women of all ages require daily intimate hygiene to keep their genital area clean
2	The vulva is susceptible to contact dermatitis. Take care to avoid contact with irritants
3	Use a hypoallergenic liquid wash with mild detergency and pH 4.2 to 5.6
4	Avoid bar soaps and bubble baths, which are abrasive and have a more alkaline pH.
5	Lactic acid-based liquids with an acidic pH may augment skin homeostasis and have been shown to be helpful in vaginal infections as an adjuvant therapy but not as a treatment
6	Vaginal douching is not recommended.
7	Wear loose-fitting cotton underwear and minimize wearing tight clothes.
8	Change underwear frequently.
9	Do not use talcum powder.
10	Use any perfumes and deodorants sparingly (after allergy testing).
11	Change tampons and sanitary pads frequently.
12	Before and after intercourse, cleanse the vulva from front to back, especially the clitoris and vulval folds
13	Do not cleanse the vulva vigorously or irrigate the vagina.
14	Use a safe method of pubic hair removal and take care to avoid sensitivity and scarring
15	Postpartum care should include frequent cleansing, drying, and using pads as necessary. Maintain dryness over any sutures. Do not use any creams.
16	Wash hands prior to children’s genital care. Use separate towels.

V. CONCLUSION

The innate defense mechanisms namely normal vaginal flora and acidic vaginal pH protect against vulvovaginal infections. Resident bacteria like Lactobacillus etc. help to maintain an acidic pH and compete with exogenous pathogens to adhere to the vaginal mucosa. Protection against infection is particularly important for pregnant women as it increases the risk of preterm delivery, neonatal meningitis, and even fetal death as well as asymptomatic bacteriuria and urinary tract infections, upper and lower genital tract infections, and postpartum endometritis. Routine washing of the vulva with plain water is beneficial to maintain cleanliness, to prevent offensive body odor and may be a useful adjunct to medical treatment. Vulvar cleansing products are not designed to treat infections rather often promote genitourinary infection by disrupting normal vaginal microbiota.

REFERENCES

- Al-Badr A, Al-Shaikh G. Recurrent Urinary Tract Infections Management in Women: A review. *Sultan Qaboos Univ Med J*. 2013 Aug;13(3):359-67. doi: 10.12816/0003256. Epub 2013 Jun 25. PMID: 23984019; PMCID: PMC3749018.
- Arab H, Almadani L, Tahlak M. The Middle East and Central Asia guidelines on female genital hygiene. *BMJ Middle East* 2011; 19: 99–106
- Aroutcheva A, Gariti D, Simon M, Shott S, Faro J, Simoes JA, et al. Defense factors of vaginal lactobacilli. *Am J Obstet Gynecol*. 2001 Aug;185(2):375-9. doi: 10.1067/mob.2001.115867. PMID: 11518895.
- Chen Y, Bruning E, Rubino J, Eder SE. Role of female intimate hygiene in vulvovaginal health: Global hygiene practices and product usage. *Womens Health (Lond)*. 2017 Dec;13(3):58-67. doi: 10.1177/1745505717731011. Epub 2017 Sep 22. PMID: 28934912; PMCID: PMC7789027.
- Cottrell BH. An updated review of evidence to discourage douching. *MCN Am J Matern Child Nurs*. 2010 Mar-Apr;35(2):102-7; quiz 108-9. doi: 10.1097/NMC.0b013e3181cae9da. PMID: 20215951.
- F. Gary Cunningham Kenneth J. Leveno. *William's Obstetrics*. 25th edition New York: McGraw-Hill Education; 2018
- Farage MA and Maibach HI. Tissue structure and physiology of the vulva. In: Farage MA and Maibach HI (eds) *The vulva: anatomy, physiology and pathology*. New York: Informa Healthcare, 2016, pp. 9–26.
- Farage MA. Vulvar susceptibility to contact irritants and allergens: a review. *Arch Gynecol Obstet*. 2005 Jul;272(2):167-72. doi: 10.1007/s00404-005-0732-4. Epub 2005 May 19. PMID: 15906051.
- Fashemi B, Delaney ML, Onderdonk AB, Fichorova RN. Effects of feminine hygiene products on the vaginal mucosal biome. *Microb Ecol Health Dis*. 2013 Feb 25;24. doi: 10.3402/mehd.v24i0.19703. PMID: 24009546; PMCID: PMC3758931.
- Fiscella K, Franks P, Kendrick JS, Bruce FC. The risk of low birth weight associated with vaginal douching. *Obstet Gynecol*. 1998 Dec;92(6):913-7. doi: 10.1016/s0029-7844(98)00325-1. PMID: 9840548.
- Goldman MB, Hatch MC, eds. *Women & health*. San Diego: Academic Press; 2000. pp 352-360.
- Hay PE, Lamont RF, Taylor-Robinson D, Morgan DJ, Ison C, Pearson J. Abnormal bacterial colonisation of the genital tract and subsequent preterm delivery and late miscarriage. *BMJ*. 1994 Jan 29;308(6924):295-8. doi: 10.1136/bmj.308.6924.295. PMID: 8124116; PMCID: PMC2539287.
- Hilier SL. Laboratory diagnosis of yeast vaginitis. In: Horowitz B, Mardh PA, eds. *Vaginitis and Vaginosis*. New York, NY: Wiley-Liss Inc; 1991. pp 121-124.
- Hillier SL, Nugent RP, Eschenbach DA, Krohn MA, Gibbs RS, Martin DH, et al. Association between bacterial vaginosis and preterm delivery of a low-birth-weight infant. The Vaginal Infections and Prematurity Study Group. *N Engl J Med*. 1995 Dec 28;333(26):1737-42. doi: 10.1056/NEJM199512283332604. PMID: 7491137.
- Holst E, Goffeng AR, Andersch B. Bacterial vaginosis and vaginal microorganisms in idiopathic premature labor and association with pregnancy outcome. *J Clin Microbiol*. 1994 Jan;32(1):176-86. doi: 10.1128/jcm.32.1.176-186.1994. PMID: 8126176; PMCID: PMC262991.
- Hull T, Hilber AM, Chersich MF, Bagnol B, Prohmmo A, Smit JA, et al; WHO GSVP Study Group. Prevalence, motivations, and adverse effects of vaginal practices in Africa and Asia: findings from a multicountry household survey. *J Womens Health (Larchmt)*.

- 2011 Jul;20(7):1097-109. doi: 10.1089/jwh.2010.2281. Epub 2011 Jun 13. PMID: 21668355.
- Hutchinson KB, Kip KE, Ness RB; Gynecologic Infection Follow-Through (GIFT) Investigators. Vaginal douching and development of bacterial vaginosis among women with normal and abnormal vaginal microflora. *Sex Transm Dis.* 2007 Sep;34(9):671-5. doi: 10.1097/01.olq.0000258435.34879.da. PMID: 17413534.
- Klebanoff MA, Nansel TR, Brotman RM, Zhang J, Yu KF, Schwebke JR, et al. Personal hygienic behaviors and bacterial vaginosis. *Sex Transm Dis.* 2010 Feb;37(2):94-9. doi: 10.1097/OLQ.0b013e3181bc063c. PMID: 19823112; PMCID: PMC2811217.
- Larsen B. Vaginal flora in health and disease. *Clin Obstet Gynecol.* 1993 Mar;36(1):107-21. doi: 10.1097/00003081-199303000-00016. PMID: 8435935.
- MacIntyre DA, Chandiramani M, Lee YS, Kindinger L, Smith A, Angelopoulos N, et al. The vaginal microbiome during pregnancy and the postpartum period in a European population. *Sci Rep.* 2015 Mar 11;5:8988. doi: 10.1038/srep08988. PMID: 25758319; PMCID: PMC4355684.
- Mignini L, Carroli G, Abalos E, Widmer M, Amigot S, Nardin JM, et al; World Health Organization Asymptomatic Bacteriuria Trial Group. Accuracy of diagnostic tests to detect asymptomatic bacteriuria during pregnancy. *Obstet Gynecol.* 2009 Feb;113(2 Pt 1):346-52. doi: 10.1097/AOG.0b013e318194f109. PMID: 19155905.
- Ness RB, Hillier SL, Richter HE, Soper DE, Stamm C, McGregor J, Bass DC, Sweet RL, Rice P. Douching in relation to bacterial vaginosis, lactobacilli, and facultative bacteria in the vagina. *Obstet Gynecol.* 2002 Oct;100(4):765. doi: 10.1016/s0029-7844(02)02184-1. PMID: 12383547.
- Olsen BE, Hinderaker SG, Lie RT, et al. The diagnosis of urinary tract infections among pregnant women in rural Tanzania;prevalence and correspondence between different diagnostic methods.*Acta Ostet Gynecol Scand/2000.Sep*
- Pete PMN, Biguioh RM, Izacar AGB, Adogaye SBB, Nguemo C. Genital hygiene behaviors and practices: A cross-sectional descriptive study among antenatal care attendees. *J Public Health Afr.* 2019 May 3;10(1):746. doi: 10.4081/jphia.2019.746. PMID: 31214303; PMCID: PMC6548999.
- Ramos BA, Kanninen TT, Sisti G, Witkin, SS. Microorganisms in the female genital tract during pregnancy: tolerance versus pathogenesis. *Am. J. Reprod. Immunol.* (2015) 73:383–9. doi: 10.1111/aji.12326.
- Rosenberg MJ, Phillips RS, Holmes MD. Vaginal douching. Who and why? *J Reprod Med.* 1991 Oct;36(10):753-8. PMID: 1956017.
- Rosenberg MJ, Phillips RS. Does douching promote ascending infection? *J Reprod Med.* 1992 Nov;37(11):930-8. PMID: 1460612.
- Rothman KJ, Funch DP, Alfredson T, Brady J, Dreyer NA. Randomized field trial of vaginal douching, pelvic inflammatory disease and pregnancy. *Epidemiology.* 2003 May;14(3):340-8. PMID: 12859036.
- Schnarr J, Smaill F. Asymptomatic bacteriuria and symptomatic urinary tract infections in pregnancy. *Eur J Clin Invest.* 2008 Oct;38 Suppl 2:50-7. doi: 10.1111/j.1365-2362.2008.02009.x. PMID: 18826482.
- Scholes D, Daling JR, Stergachis A, Weiss NS, Wang SP, Grayston JT. Vaginal douching as a risk factor for acute pelvic inflammatory disease. *Obstet Gynecol.* 1993 Apr;81(4):601-6. PMID: 8459976.
- Scholes D, H.M.Thomas,Roberts L. Pacita, Stapleton E Ann. Risk Factors for Recurrent Urinary tract Infection in young women.*The Journal of Infectious Diseases.* November 2000,182(4):1177-82

- Scholes D, Stergachis A, Ichikawa LE, Heidrich FE, Holmes KK, Stamm WE. Vaginal douching as a risk factor for cervical Chlamydia trachomatis infection. *ObstetGynecol.* 1998 Jun; 91(6):993-7. doi: 10.1016/s0029-7844(98)00095-7. PMID: 9611011.
- Sutton MY, Bruce C, Sternberg MR, Geraldine McQuillan⁴, Juliette S. Kendrick⁵, Emilia Koumans et al. Prevalence and correlates of vaginal douching among women in the United States, 2001-2002. National STD Prevention Conference 2006.
- Wakashin K. Sanitary napkin contact dermatitis of the vulva: location-dependent differences in skin surface conditions may play a role in negative patch test results. *J Dermatol.* 2007 Dec;34(12):834-7. doi: 10.1111/j.1346-8138.2007.00395.x. PMID: 18078410.
- Wølner-Hanssen P, Eschenbach DA, Paavonen J, Stevens CE, Kiviat NB, Critchlow C, et al. Association between vaginal douching and acute pelvic inflammatory disease. *JAMA.* 1990 Apr 11;263(14):1936-41. doi: 10.1001/jama.1990.03440140062032. PMID: 2313870..
- Yazici S, Demirsoy G. Urinary tract infection and genital hygiene in pregnancy. *Turk Clin Obstet Gynecol* 2009;19:241-8.