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Effect of intimate hygiene fluids on the number of Listeria monocytogenes isolated from women

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ABSTRACT

Introduction: *Listeria monocytogenes* is an etiological factor of listeriosis, widespread in the environment. The consequence of fetal infection in the second trimester of pregnancy in most cases is the death of the fetus or stillbirth.

Material and methods: In this study, 7 strains of *L. monocytogenes* isolated from the vagina of women and the reference strain *L. monocytogenes* ATCC 19111 were used. The examined strains were treated with five commercially available intimate hygiene washes. The efficacy of the wash fluids used was based on the induced decrease in the number of bacteria expressed as a logarithmic colony-forming unit (CFU) \times ml⁻¹. **Results:** The study showed that probiotic fluid (pH = 3.5) decreased the number of bacteria by an average of 4.56 log CFU \times ml⁻¹, while wash fluid intended for pregnant and puerperal women (pH = 4.0) reduced the number of *L. monocytogenes* by an average of 1.55 log CFU \times ml⁻¹. Lower bactericidal efficacy was observed in the case of wash fluids intended for everyday use. The fluid containing marigold extract and the liquid with the addition of rice proteins and arnica extract decreased the number of *L. monocytogenes* by an average of 1.11 log CFU \times ml⁻¹, and the fluid with silver and copper nanoparticles by 1.14 log CFU \times ml⁻¹.

Conclusions: Everyday use of intimate hygiene washes reduces the risk of urogenital infections in women and supports their treatment. Prevention with the use of probiotic wash solutions may reduce the number of vaginal infections caused by *L. monocytogenes*, which is especially important in the case of pregnant women because the number of patients diagnosed with listeriosis has increased.

Key words: intimate hygiene fluids, *Listeria monocytogenes*, vaginal infections, listeriosis in pregnancy Med Res J 2022; 7 (1): 24–31

Introduction

Listeria monocytogenes are Gram-positive, relatively anaerobic rods, commonly found in the natural environment [1]. In recent years, there has been an increase in the incidence of listeriosis, caused by pathogenic *L. monocytogenes*. Infections with these bacteria are common in the population of pregnant women and account for almost 27.0% of all patients with listeriosis [2]. The asymptomatic carrier of *L. monocytogenes* was also found in the vagina and gastrointestinal tract in 5–10% of people [1]. *Listeria monocytogenes* can cross the placental barrier and is a growing threat compared to other pathogens that can cause fetal damage or failure of pregnancy [3].

Listeriosis in pregnant women can be asymptomatic or there are flu-like symptoms (fever, headache, diarrhea, muscle aches). The infection can have serious consequences for the newborn [4]. Early neonatal listeriosis (the onset of infection in the uterus) is manifested by sepsis, respiratory failure, purulent conjunctivitis, and skin lesions (they appear 1.5 days after birth). The late form of neonatal listeriosis (mainly meningitis) appears a few days or weeks after birth (infection in the hospital

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or during childbirth due to L. monocytogenes being carried in the vagina) [5]. Listeriosis in newborns, children up to 4 years of age, and adults under 25 years of age occurs sporadically and more often affects girls and young women [6]. According to the CDC (2008), the incidence of listeriosis in pregnant women is 12 per 100,000 inhabitants, while in the general population, the incidence rate is 0.7 per 100,000 inhabitants. This ratio in newborns is 8.6 per 100,000 live births [7]. The estimated incidence of pregnancy-related listeriosis is from 1 to 25 cases per 100,000 births, which is up to 35% of all infections with L. monocytogenes. The incidence of listeriosis in newborns is approximately 86/10,000 live births, with high mortality (20-60%), and is one of the most common causes of neonatal meningitis [8]. In Poland, 3 cases of congenital listeriosis were reported in 2018 and 9 cases in 2019 [9]. An equally disturbing trend recorded around the world is the increase in patients with symptoms of infections of the genitourinary system [10]. The female vaginal microbiota includes mainly Lactobacillus spp., which limits the development of pathogenic microorganisms through the production of bacteriocins and hydrogen peroxide [11]. Currently, the cosmetics market offers a wide range of intimate hygiene products. Their composition includes a number of washing and probiotic substances aimed at the elimination of pathogens, including L. monocytogenes. L. monocytogenes is a pathogen resistant to temperature changes (surviving at temperatures ranging from -0.4°C to 50°C) and can multiply in a wide range of pH (from 4.4 to 9.4) [1]. Prophylaxis of infections in intimate areas with appropriately selected preparations may be an important aspect aimed at reducing cases of bacterial vaginosis among women [10, 12].

The purpose study aimed to evaluate the effect of five commercially available intimate hygiene fluids on the number of *L. monocytogenes* isolated from women.

Material and methods

Material

The study used 7 strains of *L. monocytogenes* isolated from the vagina of women (from the collection of the Department of Microbiology of Collegium Medicum im. L. Rydygier in Bydgoszcz of the Nicolaus Copernicus University in Torun) and the reference strain *L. monocytogenes* ATCC 19111. Based on our previous research, we know that all tested strains belonged to the serogroup 1/2a-3a and were susceptible to antibiotics recommended by EUCAST (European Committee on Antimicrobial Susceptibility Testing) for this species (penicillin, ampicillin, meropenem, erythromycin, and trimethoprim-sulfomethoxazole). The tested strains had genes coding for virulence factors such as *fbpA*, *hlyA*, *plcA*, *acta*, *inlB*, *plcB*, *iap*, *inlA*, *mpl*, *prfA*. The test strains were exposed to five different commercially available intimate hygiene liquids to evaluate their effectiveness:

- liquid for women during pregnancy and puerperium (pH = 4.0) — prebiotic complex composed of inulin, xylitol, glucooligosaccharides, maltodextrin, and lactitol;
- liquid with nanosilver and nanoscale a hypoallergenic product with pH = 4.5;
- liquid with calendula extract intended for daily intimate hygiene for mature women and in the period of menopause, pH = 4.5 — with provitamin B5 and lactic acid;
- liquid with arnica extract and rice proteins dedicated to hygiene and care of intimate areas in states of irritation or for everyday use;
- probiotic liquid intended for use in inflammation, bacterial vaginosis, as well as during menstruation and antibiotic therapy. The agent contains live bacteria from two species of lactobacilli (*L. casei* and *L. acidophilus*). Due to the high content of lactic acid, the pH of the liquid is 3.5.

Evaluation of bactericidal effectiveness against L. monocytogenes of selected intimate hygiene products

The *L. monocytogenes* strains were seeded on Columbia Agar with 5% sheep blood (bioMerieux) (incubation: 24h, 37°C). From the colonies grown, a suspension was prepared for each strain in a sterile saline solution (Avantor) with an optical density of 0.5 McFarland.

The prepared suspensions (1 ml) were transferred (in triplicate) to a sterile Eppendorf tube and centrifuged (5000 rpm, 5 min). The supernatant was then removed and 1 ml of the intimate hygiene fluid tested was added to the test tube. The whole was thoroughly mixed by 1-minute vortexing and left at room temperature for 20 minutes.

Next, the tubes were mixed and 100 μ l of the suspension was transferred to 900 μ l of neutralizer (Tween 80 [Sigma Aldrich] — 10 g, lecithin [Sigma Aldrich] — 1 g, histidine L [Sigma Aldrich] — 0.5 g, sodium thiosulfate [Avantor] — 2.5 g and water — 1000 ml). A series of ten-fold dilutions (from 10⁻¹ to 10⁻⁵) in sterile buffered saline solution (PBS) [BTL] was then made. From each dilution, 100 μ l was plated on Columbia Agar supplemented with 5% sheep blood (bioMerieux). After incubation (48 h, 37°C), the grown colonies were counted and expressed as CFU × ml⁻¹.

The positive control consisted of suspensions of the tested strains subjected to the entire experimental procedure, in which the intimate hygiene fluid was replaced with sterile PBS (Avantor). Negative control was intimate hygiene products without microorganisms.

Statistical analysis

The number of colonies grown on the plates was converted into the number of cells expressed as CFU × ml⁻¹. The starting number of bacteria was the mean value of the results obtained in triplicates for each strain in the control sample. The average values of the results from three repetitions in the test sample were calculated analogously. The difference in mean values for both samples was then calculated and presented as a logarithmic decrease in the number of bacteria expressed as CFU × ml⁻¹. The statistical significance of the results was tested with the Tukey test at the significance level α = 0.05.

Results

Evaluation of the bactericidal efficacy of selected intimate hygiene products

Diverse bactericidal efficacy has been demonstrated against *L. monocytogenes*, depending on the intimate hygiene fluid used and the strains of the bacilli tested.

1. Evaluation of the effectiveness of the liquid for women during pregnancy and puerperium

The initial number of bacteria tested varied depending on the strain and ranged from 6.62 log CFU \times ml⁻¹ for the LMO1 strain to 8.20 log CFU \times ml⁻¹ for LMO6 (Tab. 1). After the application of the fluid, *L. monocytogenes* number ranged from 4.40 log CFU \times ml⁻¹ for the LMO1 strain to 6.36 log CFU \times ml⁻¹ for LMO6 (Tab. 1). The observed decrease in the number of rods tested ranged from 0.75 log CFU \times ml⁻¹ for the ATCC 19111 strain to 2.22 log CFU \times ml⁻¹ for LMO1 (Tab. 1). The ATTC 19111 and LMO3 strains were statistically significantly more resistant to the hygiene fluid dedicated for pregnant and puerperal women compared to the other strains of *L. monocytogenes* tested (Tab. 1). 2. Evaluation of the effectiveness of the liquid with

nanosilver and nanoparticles

The starting bacterial cell counts were shown to range from 7.55 log CFU × ml⁻¹ for the LMO2 strain to 8.32 log CFU × ml⁻¹ for LMO6. After the application of the test agent, the number of reisolated rods decreased and ranged from 6.27 log CFU × ml⁻¹ for LMO2 to 7.45 log CFU × ml⁻¹ for LMO6 (Tab. 2). The bactericidal effect of the agent with nanosilver and nanoparticles correlated with the tested strain of *L. monocytogenes*. A decrease in the number of bacteria ranged from 0.66 log CFU × ml⁻¹ for strain ATCC 19111 to 1.90 log CFU × ml⁻¹ for strain LMO1 (Tab. 2). The reference strain *L. monocytogenes* ATCC 19111 showed the statistically significant highest resistance among all tested strains to the effect of the liquid containing nanosilver and nanoparticles (Tab. 2).

 Evaluation of the effectiveness of the liquid with marigold extract for mature women

The initial number of *L. monocytogenes* colonies ranged from 7.77 log CFU × ml⁻¹ for the LMO7 strain to 8.53 log CFU × ml⁻¹ for the LMO1 strain (Tab. 3). The liquid decreased the number of bacteria (depending on the strain tested) from 0.22 log CFU × ml⁻¹ (LMO5 strain) to 2.00 log CFU × ml⁻¹ (LMO1 strain) (Tab. 3). The strain that was statistically significantly more resistant to the tested product was LMO5, while the strain LMO1 was statistically significantly more sensitive (Tab. 3).

<i>Listeria monocytogenes</i> strain	Baseline bacterial count (log CFU×ml ⁻¹)	The number of bacteria after the action of the agent (log CFU×mI ⁻¹)	Bacteria reduction (log CFU×ml ⁻¹)
ATCC 19111	6.68	5.93	0.75°
	<i>0.07</i> *	<i>0.05</i>	0.10
LMO1	6.62	4.40	2.22 ^b
	0.06	0.14	0.06
LMO2	6.87	5.15	1.72 ^{a, b}
	0.03	0.24	0.04
LMO3	6.80	5.89	0.91°
	0.02	<i>0.15</i>	<i>0.03</i>
LMO4	7.94	6.33	1.61 ^{a, b}
	0.20	<i>0.11</i>	<i>0.21</i>
LMO5	7.72	6.34	1.38ª
	0.23	0.12	<i>0.24</i>
LMO6	8.20	6.36	1.84 ^{a, b}
	0.07	<i>0.11</i>	<i>0.08</i>
LMO7	8.19	6.23	1.96 ^{a, b}
	<i>0.04</i>	0.22	<i>0.05</i>

Table 1. The number of bacteria before and after treatment with agent for women during pregnancy and puerperium

*standard deviation; a, b, c, ... — values marked with different letters differ statistically significant

Listeria monocytogenes strain	Baseline bacterial count (log CFU×ml ⁻¹)	The number of bacteria after the action of the agent (log CFU×ml ⁻¹)	Bacteria reduction (log CFU×ml ⁻¹)
ATCC 19111	8.02	7.36	0.66ª
	0.15*	0.04	<i>0.19</i>
LMO1	8.30	6.40	1.90 ^{c, b}
	<i>0.07</i>	<i>0.04</i>	<i>0.08</i>
LMO2	7.55	6.27	1.28 ^{a, b}
	0.09	0.07	<i>0.10</i>
LMO3	8.32	7.53	0.79 ^{a, b}
	0.16	0.15	0.22
LMO4	7.99	6.99	1.00 ^{a, b}
	0.09	0.12	<i>0.09</i>
LMO5	7.76	6.72	1.04 ^{a, b}
	0.29	0.04	<i>0.34</i>
LMO6	8.22	7.45	0.77 ^{a, b}
	0.07	0.02	<i>0.09</i>
LMO7	8.00	7.10	0.90 ^{a, b}
	<i>0.06</i>	<i>0.13</i>	<i>0.06</i>

Table 2	The number	of bacteria be	fore and after the	e action of the	agent with r	hanosilver and	1 nanonarticles
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*standard deviation; a, b, c, ... — values marked with different letters differ statistically significant

Table 3. The number of bacteria before and after the action of the a	agent with	marigold extrac
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Listeria monocytogenes strain	Baseline bacterial count (log CFU×ml ⁻¹)	The number of bacteria after the action of the agent (log CFU×ml ⁻¹)	Bacteria decline (log CFU×ml⁻¹)
ATCC 19111	8.32	6.82	1.50 ^b
	0.08*	0.20	0.08
LMO1	8.53	6.53	2.00 ^d
	0.04	<i>0.11</i>	0.04
LMO2	8.10	6.88	1.22 ^{a, b}
	<i>0.12</i>	<i>0.07</i>	0.13
LMO3	8.05	6.78	1.27 ^{a, b}
	<i>0.05</i>	<i>0.05</i>	0.05
LMO4	8.14	7.31	0.83ª
	0.04	0.06	<i>0.05</i>
LMO5	8.00	7.78	0.22°
	<i>0.01</i>	0.03	0.07
LMO6	7.88	6.93	0.95 ^a
	0.04	<i>0.04</i>	<i>0.05</i>
LMO7	7.77	6.88	0.89 ^a
	0.21	<i>0.06</i>	0.24

*standard deviation; a, b, c, ... - values marked with different letters differ statistically significant

4. Evaluation of the effectiveness of the liquid with rice proteins and arnica extract

Before the use of the agent, the number of *L. mono-cytogenes* rods ranged from 8.04 log CFU \times ml⁻¹ for the LMO2 strain to 8.48 log CFU \times ml⁻¹ for the LMO6 strain (Tab. 4). After the application of the fluid tested, a decrease in these values was observed to the level of 6.76 log CFU \times ml⁻¹ (LMO3 strain) to 7.67 log CFU \times ml⁻¹ (LMO6 strain). The product decreased

from 0.72 log CFU × ml⁻¹ (LMO4 strain) to 1.68 log CFU × ml⁻¹ (LMO3 strain) bacteria number (Tab. 4).
Statistically significant, the highest resistance was demonstrated for the LMO4 and LMO7 strains (Tab. 4).
Evaluation of the effectiveness of a probiotic liquid with pH = 3.5

The initial number of bacteria tested varied, depending on the strain, from 7.70 log CFU \times ml⁻¹ (LMO1 strain) to 8.47 log CFU \times ml⁻¹ (LMO6 strain). After liquid

Listeria monocytogenes strain	Baseline bacterial count (log CFU×ml ⁻¹)	The number of bacteria after the action of the agent (log CFU×ml ⁻¹)	Bacteria decline (log CFU×ml ⁻¹)
ATCC 19111	8.28	7.11	1.17 ^{a, b}
	0.06*	0.02	<i>0.06</i>
LMO1	8.22	6.93	1.29 ^{a, b}
	0.22	0.02	<i>0.23</i>
LMO2	8.04	6.78	1.26 ^{a, b}
	<i>0.17</i>	<i>0.04</i>	<i>0.18</i>
LMO3	8.44	6.76	1.68°
	<i>0.15</i>	0.02	<i>0.15</i>
LMO4	8.36	7.64	0.72ª
	<i>0.18</i>	0.15	<i>0.29</i>
LMO5	8.35	7.17	1.18 ^{a, b}
	<i>0.13</i>	0.06	<i>0.14</i>
LMO6	8.48	7.67	0.81ª
	<i>0.05</i>	0.16	<i>0.06</i>
LMO7	8.12	7.36	0.76ª
	0.03	<i>0.18</i>	<i>0.07</i>

Table 4.	The number	of bacteria	before and	after treatment	with rice	proteins and	arnica extract
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*standard deviation; a, b, c, ... — values marked with different letters differ statistically significant

<i>Listeria monocytogenes</i> strain	Baseline bacterial count (log CFU×ml ⁻¹)	The number of bacteria after the action of the agent (log CFU×ml ⁻¹)	Bacteria decline (log CFU×ml⁻¹)
ATCC 19111	7.87	4.36	3.51 ^{c, d}
	0.49*	0.32	0.49
LMO1	7.70	5.12	2.58 ^d
	0.02	0.04	0.02
LMO2	8.03	3.65	4.38 ^{a, b}
	<i>0.02</i>	0.46	0.02
LMO3	8.09	3.40	4.69 ^{a, b, c}
	<i>0.02</i>	0.28	0.02
LMO4	8.31	3.39	4.92 ^{a, b, c}
	<i>0.04</i>	0.29	0.04
LMO5	8.11	2.43	5.80 ^{b, c}
	<i>0.0</i> 9	0.61	<i>0.09</i>
LMO6	8.47	2.36	6.11 ^c
	0.10	0.32	<i>0.10</i>
LMO7	8.14	3.56	4.58 ^{a, b, c}
	<i>0.10</i>	<i>0.32</i>	0.10

Table 5. The number of bacteria before and after the action of the agent with a probiotic pH = 3.5

*standard deviation; a, b, c, ... - values marked with different letters differ statistically significant

application, *L. monocytogenes* number ranged from 2.36 log CFU × ml⁻¹ (LMO6 strain) to 5.12 log CFU × ml⁻¹ (LMO1 strain) (Tab. 5). The number of reisolated rods was reduced by 2.58 log CFU × ml⁻¹ (LMO1 strain) to 6.11 log CFU × ml⁻¹ (LMO6 strain) (Tab. 5). The LMO1 strain was statistically significantly more resistant to the action of probiotic fluid with pH = 3.5, compared to the other strains of *L. monocytogenes* tested, while the LMO6 strain was statistically significantly the most sensitive (Tab. 5).

In the negative control, no microbial growth was detected, confirming the microbiological purity of the intimate hygiene products tested.

Comparison of the effectiveness of the intimate hygiene products tested against Listeria monocytogenes

The highest antimicrobial efficacy was demonstrated for the probiotic liquid, which decreased the number of



Figure 1. The mean values of the decrease in the number of bacteria after the application of the measures (a, b — values marked with different letters differ statistically significant [$p \le 0.05$]); *standard deviation

bacteria by an average of 4.56 log CFU \times ml⁻¹ (Fig. 1). This value was statistically significantly higher than the effectiveness of other fluids tested. The effectiveness of other intimate hygiene liquids did not differ significantly statistically. They reduced from 1.04 log CFU \times ml⁻¹ (liquid with nanosilver and nanocooper) to 1.55 log CFU \times ml⁻¹ of *L. monocytogenes* number (liquid for pregnant and puerperal women) (Fig. 1).

Discussion

Listeria monocytogenes is hazardous due to its presence in the environment and food. Listeriosis in pregnant women is a serious threat to the life of the fetus [13]. As prophylaxis, elimination of certain food (e.g., smoked fish) during pregnancy is recommended to avoid *L. monocytogenes* infection. The purpose of the study was to evaluate the effect of various intimate hygiene preparations on *L. monocytogenes* strains. The results of our research expand the topic of the effectiveness of intimate hygiene products in the prevention of infections with *L. monocytogenes* in women. Features such as the selection of active ingredients and pH affect the antibacterial properties of the preparations.

All intimate hygiene products used in the study reduced *L. monocytogene* growth. On the contrary, Wysocka-Lipiska et al. [14] demonstrated no antibacterial properties of intimate wipes. The low antibacterial effectiveness of wipes was explained by a small amount of the active solution in the unit package and its composition compared to intimate hygiene liquids. Therefore, it can be concluded that the use of intimate hygiene fluids is more beneficial in the prevention of genital tract infections in women than the use of intimate wipes.

The study included five commercially available intimate hygiene products, differing in composition and purpose. The own study showed the highest antibacterial effectiveness of the probiotic fluid. The lowest effectiveness was noted in the case of the fluid dedicated to women during pregnancy and puerperium. The difference in the effectiveness of these agents was most likely due to the pH variation. In turn, Bordes et al. [4] showed that vaginal pH had an impact on L. monocytogenes survival, and acidic vaginal pH (4.2) reduced the number of pathogenic bacilli. Researchers also showed that at pH 6.5 all L. monocytogenes strains were able to grow, therefore vaginal colonization with L. monocytogenes strains is possible only in the case of pH increase [4]. The three consecutive intimate hygiene fluids used in the study were intended for daily use and probably, for this reason, these agents in the experiment showed very similar efficacy. Silver and copper nanoparticles added to one of these products have been proven to be highly bactericidal. Chen et al. [15] conducted research on the antibacterial effectiveness of gels with the addition of silver nanoparticles intended for use in the treatment of bacterial vaginosis. They used three species of bacteria in the study: Staphylococcus aureus, Escherichia coli, and Pseudomonas aeruginosa. Chen et al. [15] showed that S. aureus bacteria, compared to other species, were characterized by the highest sensitivity to the gel tested, while E. coli bacteria were the most resistant to the agent [15]. In turn, Tamayo et al. [16] revealed a decrease in the number of L. monocytogenes of approximately 33% after 6 hours of exposure to a gel with a content of 1% silver nanoparticles in a polyethylene nanocomposite. After 24 hours of exposure to the gel with the 5% addition of nanosilver, no L. monocytogenes were found [16]. In our study, the liquid with the addition of silver and copper nanoparticles reduced the number of L. monocytogenes by 1.04 log CFU \times ml⁻¹ on average, that is, by more than 90%. The third liquid was enriched with calendula extract, which, due to essential oils, also has a bactericidal effect and protects against excessive drying of the mucosa. There are no detailed data in the literature on the effectiveness of individual active ingredients contained in intimate hygiene liquids tested. In our study, agents in this group, regardless of the variety of active ingredients, showed very similar effectiveness. The pH of these preparations was 4.5 and the regulator of this parameter was lactic acid. Bahamondes et al. [17] investigated the effect of a high lactic acid intimate hygiene soap with pH = 3.5 on the number of recurrences of genital tract infections among women treated with metronidazole. They showed that the product reduced the number of recurrent vaginal infections in women caused by bacteria and Candida spp. [17]. According to Biaobrzeska [18], the use of intimate hygiene liquids is also an important element in the prevention of urinary tract infections in patients after kidney transplantation [18]. The research by Bruning et al. [19], assessing the impact of the daily use of intimate hygiene liquids with lactic acid (pH = 4.2), showed that liquids do not have a significant impact on the qualitative and quantitative composition of the external microbiome of the vagina and vulva [19].

Recently, the popularity of intimate hygiene products enriched with live lactobacilli has increased. The first reports of the use of probiotic products as supporting elements in the treatment of bacterial vaginosis appeared in the 1930s. This issue was extended in the work by Pawowska [20], which showed that the use of available vaginal probiotics on the market alleviated the symptoms of bacterial vaginosis and limited the development of candidiasis. Furthermore, it has been shown that the greater the variety of lactobacilli strains contained in the probiotic, the wider spectrum activity of the product [20]. The intimate probiotic hygiene fluid used in our work contained bacteria from two species: L. casei and L. acidophilus. Based on the results of ourresearch, it can be concluded that the addition of live lactobacilli cultures enhances the anti-Listeria

properties of the tested probiotic agent. The great interest of researchers is focused on oral gynecological probiotics. The most commonly used strains (with proven effect) are *L. plantarum* and *L. rhamnosus*. Oral probiotics are an interesting modern therapeutic option to restore the natural vaginal microbiota and prevent recurring infections. The results confirming their effectiveness were presented by Harasim-Dylak et al. [21].

Conclusions

The use of intimate hygiene fluids reduces the risk of urogenital infections in women. The use of preparations enriched with probiotic strains may reduce the number of infections caused by *L. monocytogenes*, which is particularly important in the case of pregnant women and the increasing number of patients diagnosed with listeriosis in recent years.

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References

- Jurkiewicz A, Oleszczak-Momot W. Listeria monocytogenes jako problem zdrowia publicznego. Med Og Nauk Zdr. 2015; 21(1): 29–32.
- Schlech FW. Epidemiology and clinical manifestations of Listeria monocytogenes infections. In: Fischetti V, Novick R, Ferretti J, Portnoy D, Rood J. ed. Gram-Positive Pathogens Vol. 2. ASM Press, Washington 2006: 601–608.
- Boroń-Kaczmarska A. Zakażenia u ciężarnych czy TORCH nadal jest dominujący? Zakażenia XXI w. 2019; 2(3): 107–113.
- Borges SF, Silva JGL, Teixeira PCM. Survival and biofilm formation of Listeria monocytogenes in simulated vaginal fluid: influence of pH and strain origin. FEMS Immunol Med Microbiol. 2011; 62(3): 315–320, doi: 10.1111/j.1574-695X.2011.00815.x, indexed in Pubmed: 21569122.
- Posfay-Barbe KM, Wald ER, et al. Listeriosis. Semin Fetal Neonatal Med. 2009; 14(4): 228–233, doi: 10.1016/j.siny.2009.01.006, indexed in Pubmed: 19231307.
- Krajowy Ośrodek Referencyjny ds. Diagnostyki Bakteryjnych Zakażeń Ośrodkowego Układu Nerwowego: Listerioza w Polsce w latach 2011 - 2015 Warszawa, 17.03.2016.
- Centers for Disease Control and Prevention (CDC). Preliminary Food-Net data on the incidence of infection with pathogens transmitted commonly through food--10 states, 2007. MMWR Morb Mortal Wkly Rep. 2008; 57(14): 366–370, indexed in Pubmed: 18401330.
- Madjunkov M, Chaudhry S, Ito S. Listeriosis during pregnancy. Arch Gynecol Obstet. 2017; 296(2): 143–152, doi: 10.1007/s00404-017-4401-1, indexed in Pubmed: 28536811.
- NIZP-PZH. Choroby zakaźne i zatrucia w Polsce. Warszawa (Poland): Narodowy Instytut Zdrowia Publicznego – Państwowy Zakład Higieny. 2019.
- Atassi F, Brassart D, Grob P, et al. Lactobacillus strains isolated from the vaginal microbiota of healthy women inhibit Prevotella bivia and Gardnerella vaginalis in coculture and cell culture. FEMS Immunol Med Microbiol. 2006; 48(3): 424–432, doi: 10.1111/j.1574-695X.2006.00162.x, indexed in Pubmed: 17059467.

- Bodaszewska-Lubas M, Brzychczy-Wloch M, Gosiewski T, et al. Antibacterial activity of selected standard strains of lactic acid bacteria producing bacteriocins--pilot study. Postepy Hig Med Dosw (Online). 2012; 66: 787–794, doi: 10.5604/17322693.1015531, indexed in Pubmed: 23175332.
- Chen Y, Bruning E, Rubino J, et al. Role of female intimate hygiene in vulvovaginal health: Global hygiene practices and product usage. Womens Health (Lond). 2017; 13(3): 58–67, doi: 10.1177/1745505717731011, indexed in Pubmed: 28934912.
- Sieroszewski P, Bober Ł, Kłosński W. Zakażenia podczas ciąży. Perinatol Neonatol Ginekol. 2012; 5(2): 65–84.
- Wysocka-Lipińska N, Tkachenko H, Kurhaluk N. Ocena skuteczności chusteczek antybakteryjnych. Słupskie Prace Biologiczne. 2012; 9: 181–196.
- Chen M, Pan X, Wu H, et al. Preparation and anti-bacterial properties of a temperature-sensitive gel containing silver nanoparticles. Pharmazie. 2011; 66(4): 272–277, indexed in Pubmed: 21612154.
- Tamayo LA, Zapata PA, Vejar ND, et al. Release of silver and copper nanoparticles from polyethylene nanocomposites and their penetration into Listeria monocytogenes. Mater Sci Eng C Mater Biol Appl. 2014; 40: 24–31, doi: 10.1016/j.msec.2014.03.037, indexed in Pubmed: 24857461.

- Bahamondes MV, Portugal PM, Brolazo EM, et al. Use of a lactic acid plus lactoserum intimate liquid soap for external hygiene in the prevention of bacterial vaginosis recurrence after metronidazole oral treatment. Rev Assoc Med Bras (1992). 2011; 57(4): 415–420, doi: 10.1590/s0104-42302011000400015, indexed in Pubmed: 21876923.
- Białobrzeska P. Profilaktyka zakażeń układu moczowego u pacjentów po przeszczepie nerki. Forum Nefrol. 2011; 4(3): 266–271.
- Bruning E, Chen Y, McCue KA, et al. A 28 Day Clinical Assessment of a Lactic Acid-containing Antimicrobial Intimate Gel Wash Formulation on Skin Tolerance and Impact on the Vulvar Microbiome. Antibiotics (Basel). 2020; 9(2), doi: 10.3390/antibiotics9020055, indexed in Pubmed: 32024047.
- Pawłowska K. Wpływ liczby szczepów bakterii Lactobacillus na skuteczność probiotyku ginekologicznego. http://docplayer.pl/6108076--Wplyw-liczby-szczepow-bakterii-lactobacillus-na-skutecznosc-prob iotyku-ginekologicznego.html (19.06.2021).
- Harasim-Dylak A, Roguska M, Maździarz A. Skuteczność preparatu Trivagin w przywróceniu i utrzymaniu prawidłowego ekosystemu pochwy u kobiet leczonych z powodu nawracającej waginozy bakteryjnej. Curr Gynecol Oncol. 2011; 9(4): 245–252.