BACTERIAL VAGINOSIS Presence in Sexually Active Women in Tuzla canton Area

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Abstract

The goal of our research was to determine the presence of bacterial vaginosis in sexually active women in Tuzla Canton area. Diagnosis determination for bacterial vaginosis was conducted on the basis of three out of four internationally accepted criteria according to Amsel and isolation and identification of Gardnerella vaginalis (G. vaginalis) by standard microbiological procedures. Bacterial vaginosis was diagnosed in 20,5 % (41/200) women who asked for gynaecologist's help due to their personal discomfort, since significantly higher percentage of diagnosed bacterial vaginosis of 48,80% (41/84) was determined in women with personal discomfort typical for this disease. All relevant factors, according to available literature, for genesis of bacterial vaginosis were processed in this research. In respect to the obtained outputs, bacterial vaginosis is significantly more frequent occurrence in women who are not married, since the number of sexual partners, the time of the first sexual intercourse, the use of intrauterine contraceptive device and smoking do not cause the genesis of bacterial vaginosis. According to Nugent, an increased vaginal discharge with unpleasant odour after sexual discourse, its pH>4,5, a positive amino odour test, an occurrence of clue cells in a direct microscopic concoction of vaginal discharge and assessment of the state of vaginal flora for bacterial vaginosis are significantly more frequent occurrences in women with individual discomforts. It was proved that G. vaginalis is a dominant micro organism in 95% of women with clinical signs of vaginosis although it was isolated from vaginal discharge in 40 to 50% of healthy women. In our research, G. vaginalis was isolated in 63,41% of examined women with all signs of bacterial vaginosis, in 36,59% of examined women with one or more clinical signs of bacterial vaginosis and in 2,58% of examined women of control group without clinical signs.

KEY WORDS: bacterial vaginosis, Gardnerella vaginalis

INTRODUCTION

An increased vaginal discharge is one of the most frequent symptoms of female genital tract. The most recent research have shown that 96% of all increased vaginal discharges or infections result from five states: bacterial vaginosis from 4 to 64%; vulvovaginal candidiasis from 28 to 37%; cervicitis caused by Chlamydia trachomatis, Neisseria gonorrhoea, Herpes simplex virus type 2, from 2 to 20%; trichomonas colpitis in around 10% of women and finally superfluous but normal discharge (1). Bacterial vaginosis is a chronic or relapsing syndrome connected to unidentified agents, and pathogenesis is polymicrobial and multicausal. It was internationally defined in 1984 as "replacement of vaginal Lactobacillus with specific group of bacteria, because of which, the characteristics of vaginal discharge are changed" (3).

Clinical signs and symptoms of bacterial vaginosis are: an increased grey-white vaginal discharge with consistency similar to milk and with strong odour of fish. Bacterial vaginosis is associated with an increased risk from several pathological states including post surgery infections which arose after hysterectomy, postabortal pelvic inflammatory disease and plasma cell endometritis (4, 5, 6). In expectant mothers, bacterial vaginosis is associated with the presence of fetal fibronectin in cervikovaginal discharge during the second and the third trimester by which the risk of clinical chorioamnionitis, neonatal sepsis, preterm births has been increased for 16 times. One of the consequences of bacterial vaginosis might be cervical neoplasia due to oncogene potential of nitrosamine which is produced by anaerobic bacteria in causal relation to bacterial vaginosis (7).

Women who have had more than five sexual partners, users of intrauterine devices, smokers and persons who are diagnosed with some other sexually contagious disease suffer more frequently from bacterial vaginosis (8, 9).

Bacterial vaginosis arises as a result of normal vaginal flora exchange (*Lactobacillus*) with mixed bacterial flora which consists of *Gardnerella vaginalis*, anaerobic bacteria (*Prevotella bivia*, *Prevotella disiens*, *Prevotella species*, *Peptostreptococcus spp.*, *Mobiluncus species*) and *Mycoplasme hominis*. It has been proved that *Gardnerella vaginalis* (*G.vaginalis*) is a dominant micro organism in 95% of women with clinical signs of vaginosis although it has been isolated from vaginal discharge in 40 to 50% of healthy women.

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The occurrence of frequent relapses which appears in 20 to 30% of women within 3 months after treatment is explained by reinfection with another biotype of *G. vaginalis* which is different from genuine biotype or as a consequence of improper therapy (10). Pathogenesis of this more and more frequent clinical syndrome has not been cleared so far. The majority of authors consider hormonal factors as those which have a major role in pathogenesis of bacterial vaginosis, because that state hits only the women of reproductive age (7).

According to other authors, bacterial vaginosis is the result of changed vaginal antibiosis or antagonism between micro organisms which exist in vagina. Antibiosis is under the influence of vaginal discharge pH lowered by lactobacilli's production of acid products.

The diagnosis of bacterial vaginosis is clinically and laboratory established. It is necessary to satisfy at least three out of four accepted criteria in order to reach a clinical diagnosis: acidity of vaginal discharge higher than 4,5; homogeneous, adherent discharge; positive amino odour test; the presence of clue cells (11). Vaginal discharge pH is the most sensitive, but the least specific characteristic of Amsel's criteria. There are many factors such as vaginal rinsing, sexual intercourse, bleeding that can influence the value of pH. Acidity of vaginal discharge lower than 4,5 rejects the possibility of the presence of bacterial vaginosis.

Vaginal discharge whose odour is unpleasant is the only clinical symptom stated by the women with bacterial vaginosis. In laboratory conditions, it can be proved by amino odour test, which also can be positive in women with infection caused by Trichomonas vaginalis. Clue cells are flat epitel cells whose surface is covered by bacteria of *G. vaginalis* morphotype. They are the most specific and authentic indicator of bacterial vaginosis. In native preparation of vaginal discharge those cells can be seen as peeled, mature, superficial vaginal cells whose edges are wrinkled like ramparts and granulated cytoplasm due to adherence of G. vaginalis. But all the cells are not as clue cells comprised of adherent G. vaginalis. Sometimes, they consist of curved rod-shaped bacteria compatible with Mobiluncus spp. which hang on those cells and whose shape is similar to comma. Those cells are called commacells (12). The most often, clue cells can be found in vaginal swab, but also in sperm and urethral discharge. They are the best diagnosed in native preparation of vaginal discharge, since their Gram stain test gives more false positive results. For diagnosis of bacterial vaginosis it is necessary to find 5 to 20% clue cells relative to the whole number of epitel cells in preparation coloured by Gram, on the contrary to only one cell in 20 fields of native preparation in case of increase of 400 times. In the recent time, the assessment of the state of vaginal flora has been applied by the examination of a direct microscopic preparation which is coloured by Gram. By the system of counting of specific micro organisms whose morphotype is Gardnerella, Prevotella, Mobiluncus and Lactobacillus, the state of vaginal flora can be assessed as: normal bacterial flora, changed bacterial flora and bacterial flora which tells us about the presence of bacterial vaginosis (12).

G. vaginalis cultivation on human two-layer Tween blood agar is used as microbiological laboratory method to diagnose bacterial vaginosis (13). The minimum diagnostical criteria for identification of *G. vaginalis* are: the occurrence of β hemolysis on two-layer Tween blood agar, typical morphology of colonies and typical morphology of micro organisms in preparations coloured by Gram, negative catalase test and positive test of hippurate hydrolysis.

Additional differential diagnostical characteristics of *G. vaginalis* are negative mannitol fermentation and occurrence of inhibition zones on the nutritious agar with 50 micrograms of metronidazole and 5 micrograms of trimethoprim. The enzyme tests (ELISA), molecular biological tests (DNA-DNA hybridization), direct or indirect immunofluorescence tests (DIF and IIF) with polyclonal antibodies are used for specific detection of *G. vaginalis* (14).

The goal of our study was to:

1. Determine frequency of bacterial vaginosis findings among the population of sexually active women in Tuzla Canton area.

2. Determine frequency of isolation of *G. vaginalis* from vaginal swab among:

- women with clinical signs of bacterial vaginosis

- women without clinical signs of bacterial vaginosis

MATERIAL AND METHODS

The research was conducted at the Department for Microbiology and at the Gynaecological and Obstetric Department at the Tuzla University Clinical Centre, as well as at the Clinic for Women Health Care at the Tuzla Health Centre. A total of 200 women aged from 20-51 are included in the prospective study. Based on clinical examination and presence of one or more signs of internationally accepted Amsel criteria, the examinees are divided into the test and the control group.

- Test group consisted of 84 examinees with one or several Amsel signs of bacterial vaginosis. - Control group consisted of 116 examinees without any Amsel signs of bacterial vaginosis. Each examinee, besides registering personal and anamnestic data relevant for setting diagnosis of bacterial vaginosis, was subjected to clinical and microbiological examination. Clinical examination was performed by a specialist gynaecologist in gynaecological policlinic and the samples for microbiological examination were taken in the Institute for Microbiology. The clinical diagnosis of bacterial vaginosis was determined according to the set of internationally accepted Amsel criteria. The look and odour of vaginal discharge were analyzed and described in every examinee. The acidity of vaginal secretion was established, the amino odour test and microscopic examination of native preparation of vaginal secretion to the presence of clue cells were completed. - The acidity of vaginal secretion was determined by colour-indicating sheets of paper which were carefully put in lateral vaginal vault. The values of pH higher than 4,5 were significant for diagnosis of bacterial vaginosis.

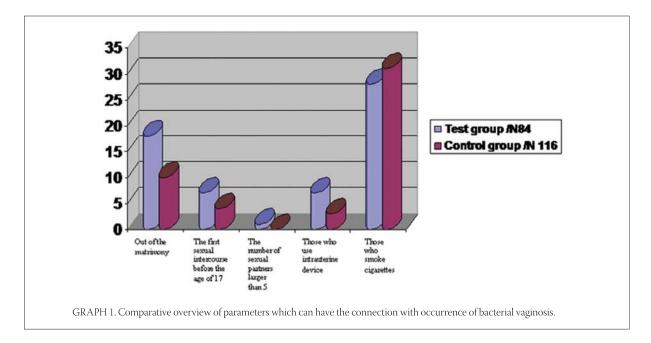
- The amino odour test was performed by putting one drop of vaginal secretion in a 10% drop of potassium alkali mixture. The discharge of strong fishy odour determined a positive amino odour test. - The microscopic examination of native preparations of vaginal discharge, the finding of only one clue cell in 20 fields of microscopic preparation during the increase of 400 times was taken as relevant for determination of bacterial vaginosis diagnosis. Three swabs of vaginal discharge were taken for microbiological examination, and they were tested as follows: 1. A direct microscopic preparation was made from the material taken by one swab, using Gram staining method. Using the system for counting characteristic microorganisms of Gardnerella, Prevotella, Mobiluncus and Lactobacilus morphotype, the assessment of the condition of vaginal flora was conducted following Nugent method (12). The test results values from 0 to 3 is marked as normal vaginal flora, from 4 to 6 as changed vaginal flora and from 7 to 10 signify the presence of bacterial vaginosis.

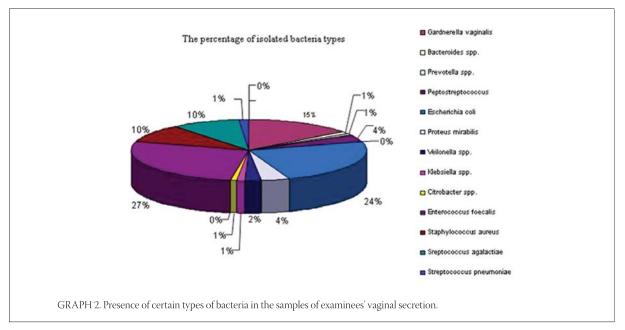
2. The second sample was cultivated based on commercial selective medium for *G. vaginalis* produced by "Sanofi" Pasteur. After the cultivation for 48 hours at 37°C, in atmosphere enriched with CO2, the identification of grown colonies was conducted through application of standard microbiological methods. Isolated types of G. *vaginalis* were identified through the occurrence of β hemolysis on human two-layer Tween blood agar, typical morphology of colonies and typical morphology of cells in preparations coloured according to Gram test, negative catalase test and positive test of hippurate hydrolysis. Test results are presented both graphically and in tabular form. χ^2 tests were used for statistic data processing, student t-test (Computer application SPSS for Windows release). Rate variations χ^2 > 3,804 we considered statistically significant.

RESULTS

The results of parallel anamnesis data of examinees from both the test and control group which are relevant for occurrence of bacterial vaginosis

Among the test group examinees 78,57% (66/84) were in matrimony, since 21,42 % (18/84) were single. In the control group, 91,37% (106/116) lived in matrimony, and 8,62% (10/116) were single. Comparing the frequency of becoming ill with bacterial vaginosis among the examinees who lived in matrimony and those who did not, it was noticed significantly higher frequency of becoming ill with bacterial vaginosis at the test group examinees ($\chi^2 = 5,617$; p<0,05). The first sexual intercourse before the age of 17 had 8,33% (7/84) test group examinees and 3,44% (4/116) control group examinees. It was determined by t-test and χ^2 test that there is not any significant difference (χ^2 = 1,369; p<0,05) in becoming ill with bacterial vaginosis compared to the age when the examinees had their first sexual intercourse. Until the day of examination, 1,19% (1/84) of test group examinees had had five or more partners during their sexual life. There were not the examinees in the control group who had had five or more sexual partners. It was determined by t-test and χ^2 test that there is not any significant difference ($\chi^2 = 0.026$; p<0,05) in becoming ill with bacterial vaginosis compared to the number of the examinees' sexual partners. During the examination of 84 examinees with one or more clinical signs of bacterial vaginosis, 25 of them or 29,76% stated that they had used contraceptive devices. 28% out of that group (7/25) uses intrauterine contraceptive device. Thirty one or 26,72% examinees in a group of 116 without any signs of bacterial vaginosis stated that they used contraceptive devices, and 9,67% (3/31) used intrauterine device. It was determined by t-test and χ^2 test that there is not any significant difference $(\chi^2 = 2,041; t = 1,9 \text{ for } p<0,05)$ in becoming ill with bacterial vaginosis compared to the use of intrauterine device as protection against unwanted pregnancy. Taking personal anamnesis from the test group examinees it was established that 33,33% (28/84) were smokers, and that the number of smokers in the control group equals 26,72% (31/116). It was determined by t-test and χ^2 test that there is not any significant difference ($\chi^2 = 0.730$) among the test and control group examinees in becoming ill with bacterial vaginosis compared to the smoking cigarettes (Graph 1).





Enterococcus foecalis was isolated from the examinees' samples of vaginal discharge in 26,95%. Escherichia coli were isolated in 23,91%, *G. vaginalis* in 14,78%, since the other bacteria were isolated in a lower percentage as it is shown in Graph 2.

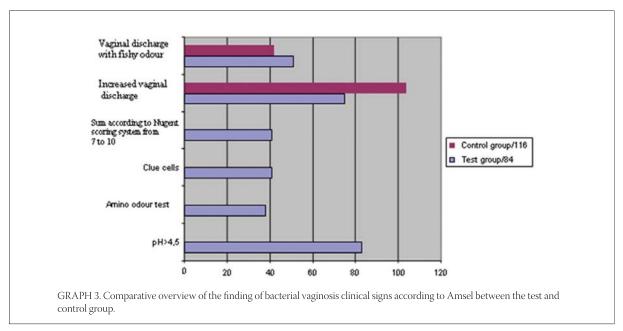
Among 200 examined samples of vaginal discharge, Candida albicans was isolated in 41 cases or in 20,5%, since Trichomonas vaginalis was found in 7 cases or in 3,5% of the samples.

An increased vaginal discharge was reported by 89,28% (75/84) of the test group examinees and by 89,65% (104/116) of the test group examinees. It was determined by χ^2 test that there is not any significant difference (χ^2 = 0,022; for p<0,05) in the occurrence of an increased vaginal discharge among the test and control group examinees. χ^2 test determined the significant difference in the occurrence of an increased vaginal discharge in the test group examinees after a sexual intercourse. The increased vaginal discharge after a sexual intercourse was noticed in 33,33% (25/75) of the test group examinees and 15,1% (18/116) of the control group examinees. After sexual intercourse, the vaginal discharge in test group examinees acquired an unpleasant fishy odour in 68% (51/75) cases, since that occurrence was noticed in 46,66% (42/90) of control group examinees which represents statistically very remarkable difference (χ^2 = 6,727).Comparing the attributes of the vaginal discharge between test and control group examinees, the statistically significant difference (p<0,01) was found only in the occurrence of an increased vaginal discharge with unpleasant odour after the sexual intercourse. The examination of the vaginal

discharge acidity in the control and test group examinees determined that 98,80% (83/84) of test group examinees and 0/116 of the control group examinees had an increased pH>4,5. χ^2 test determined that there was highly significant difference of vaginal discharge pH values between the control and test group ($\chi^2 = 191,881$). The positive Amino odour test was noticed in 45,23% (38/84) of the test group examinees, since the control group examinees had negative test (p<0,001; $\chi^2 = 61,879$). Clue cells were found in a direct microscopic preparation coloured according to Gram in 48,80% (41/84) of the test group examinees, since those cells were not found in the control group examinees. χ^2 test determined that there was highly significant difference $(\chi^2 = 68,256)$ in the finding of characteristic microscopic preparation of the vaginal discharge between the control and test group examinees (Graph 3).

The examinees of the test group, in various percentages, had one or more signs from the set of internationally accepted Amsel criteria: 44,04% (37/84) had one sign, 8,33% (7/84) had two signs, 11,90% (10/84) had three signs and 35,71 (30/84) had four out of four signs. Considering the frequency, the following clinical signs of bacterial vaginosis occurred: an increased pH 98,80%, clue cells 48,80%, characteristic finding of direct microscopic preparation according to Nugent 48,80% and positive amino odour test 45,23% (Graph 4).

The isolation of *G. vaginalis* is directly dependant on the number of expressed clinical signs of bacterial vaginosis. Significantly larger number ($\chi^2 = 15,469$) was isolated in the persons with all four clinical signs expressed. *G. vaginalis* was not isolated from the vaginal



secretion samples in examinees with only one clinical sign which is significantly less (χ^2 = 20,662) compared to the examinees with more signs (Table 1).

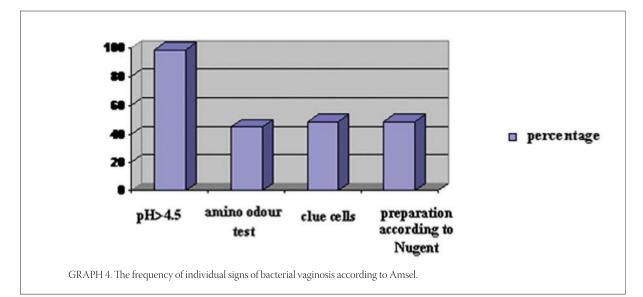
Number of clinical signs	Number of examinees	Number of <i>G.</i> <i>vaginalis</i> isolates	χ² test =/>3,840
One sign	38	0	20,662
Two signs	7	3	0,077
Three signs	10	5	0,012
Four signs	29	26	15,469
Total	84	34	

TABLE 1. The connection between *G. vaginalis* isolation and the presence of clinical signs of bacterial vaginosis.

G. vaginalis was isolated in 63,41% of the examinees with all signs of bacterial vaginosis, in 36,59% of the examinees with one or more clinical signs of bacterial vaginosis and in 2,58% of the examinees without any clinical signs.

DISCUSSION

Bacterial vaginosis or vaginal bacteriosis is polymicrobic clinical syndrome. Around 50% of women are asymptomatic with microbiological finding which suggests bacterial vaginosis. Due to that data and effects of bacterial vaginosis on the women health the timely and correct diagnosis of bacterial vaginosis is necessity. Although this disease was defined as a separate entity in 1955, it has been little known about its spread in the world and in our country. According to the results of the examinations in the USA, bacterial vaginosis is present in 26-37% of women (15, 16), and this range in European countries is from 4 to 37% (8). The detailed research of the bacterial vaginosis issue was performed in Croatia by Tomljanovic who examined various groups of women: secondary school students, women without subjective discomforts, women with



subjective discomforts typical for bacterial vaginosis, expectant mothers and women with neoplasia of cervix. The lowest percentage of 4% was found in secondary school students and the highest level of 64% in women with typical signs of bacterial vaginosis, in women without symptoms (15-25%), then expectant mothers (15,8%) and women with cervical neoplasia (48,8%) (9, 17). According to the literature information, the similar researches which could help us to assess health importance of this disease have not been performed in our country so far. In our research, bacterial vaginosis was diagnosed in 20,5% of women who, due to subjective discomforts, were examined in the Health centre for women health protection in Tuzla, which is in accordance to the literature data. However, the larger percentage of diagnosed bacterial vaginosis (40,8%) was found in women with subjective discomforts typical for this disease. Pathogenesis of bacterial vaginosis and the ways of its transmission are still unknown. There are contradictory opinions about sexual way of bacterial vaginosis transmission. The thesis that bacterial vaginosis is not sexually transmitted disease is supported by Bump and associates (18). They support their attitudes by data that they diagnosed bacterial vaginosis in 15% of sexually active women and in 12% of adolescents without sexual experience. On the other side, Barbone and associates of the opinion that bacterial vaginosis is sexually transmitted because their examinations proved that sexually active women become ill more frequently (19). The majority of authors agree that women with bacterial vaginosis start their sexual life significantly earlier than women without bacterial vaginosis (8, 20, 9, 21). They presume that earlier sexual activity leads to larger number of sexual partners and, according to that, to the higher possibility of being infected with a new pathogen or, in case of bacterial vaginosis, with a new biotype of G. vaginalis. The first sexual intercourse before the age of 17 we were evidenced in a negligible number of examinees, only 5,5% of them (11/200). Very low percentage of them, which is 1,5% (3/200), had bacterial vaginosis. Among the test group women who became sexually active before the age of 17, bacterial vaginosis was diagnosed in 3,57% (3/84) cases. This importantly differs from data obtained by Tomljanovic who found that 32% of examinees with bacterial vaginosis became sexually active before the age of 17. This relation between the occurrence of bacterial vaginosis and the number of sexual partners could not be confirmed in our research due to the fact that only 1,19% of examinees claimed to have five or more sexual partners in their sexual life. The explanation for a low percentage of the examinees can be find in different

traditional measures of women behaviour in our society. Some of those measures are definitely those related to numerous restrictions in the field of sexual behaviour. According to the data of numerous authors, bacterial vaginosis is more frequently diagnosed in the users of intrauterine devices (IUD) than in those who use other methods of contraception or do not use contraception at al. The data obtained by Tomljanovic and associates (9) shows that bacterial vaginosis is more often diagnosed in the users of intrauterine devices (47,7%) than in the users of pills (20%) and that the frequency of bacterial vaginosis is significantly low (2,7%) in women whose sexual partners used condoms. Contrary to the presented data, the results obtained by the most comprehensive study about bacterial vaginosis conducted in Sweden negate any corelation between the use of intrauterine device and the occurrence of bacterial vaginosis (22).Considering the results of our research, only 29,76% of examinees used contraceptive devices as follows: 28% of them used IUD, 52% of them used pills and 16% of examinees' sexual partners used condoms. It was not determined statistically significant difference in the use of IUD by examinees with bacterial vaginosis and those in the control group. Determination of bacterial diagnosis is a very complex work in which gynaecologists and microbiologist should cooperate and be involved. The value of findings of individual clinical signs in diagnostics of bacterial vaginosis is still under discussion and is not unified (23). The increased vaginal discharge with unpleasant odour, according to Thomason and associates, appears in 80% of women with bacterial diagnosis (8). Contrary to his claims, Tomljanovic finds the increased vaginal discharge in 38,6% of examinees and 33,2% of them are those with bacterial vaginosis and 66,8% of them are without vaginosis (9). In our research, such characteristics of secretion were identified in 44,07% of the examinees with typical signs of bacterial vaginosis and in 18,96% of the examinees with discomforts untypical for bacterial vaginosis. Only the presence of the increased vaginal discharge, according to the results from literature and those obtained by us, does not have diagnostic importance, but characteristically changed vaginal discharge, according to our research and the researches of other authors (8, 23), can be considered as an important diagnostic finding in determination of clinical diagnosis of bacterial vaginosis. According to Amsel and associates (10), the pH value of vaginal discharge increases during bacterial vaginosis. Taylor et al. (24) found that 95% of women had the increased vaginal discharge with unpleasant odour and high pH values. In the group of the examinees with signs of bacterial vaginosis, the increased pH had

98,80% of the examinees, since 97,56% of them had the increased pH above 5 in the group of the examinees with typical microscopic preparation. Therefore, considering the data above, we can conclude that pH determination of vaginal discharge has an important diagnostic value. The pH determination of vaginal discharge is a useful method for exclusion of bacterial vaginosis due to high negative predictive value of 94,2%. Chen and associates (25) first described the presence of diamines in vaginal liquid in women with vaginal diagnosis as products of anaerobic bacteria metabolism (fishy odour). They found the presence of diamine in vaginal secretion in 88% of women with diagnosed bacterial vaginosis and in 10% of women without diagnosed bacterial vaginosis. Our researches are in the accordance with the stated claims. In our research, a positive amino odour test was identified in 75,60% of the examinees with diagnosis of bacterial vaginosis and in 45,23% of the examinees with the signs of bacterial vaginosis. The examinees without discomforts typical for bacterial vaginosis did not have a positive amino odour test. According to the results of the majority of the authors, the clue cells finding is the most sensitive, the most specific and the most authentic indicator of bacterial vaginosis (23, 10, 26). According to Larsson and associates (26), bacterial vaginosis is in a good co-relation with the clue cells finding in a native preparation of vaginal discharge. They found those cells in 96,2% of women with vaginosis. All patients with such finding had also the finding of isolated G. vaginalis. In our study, clue cells were found in: 100% of the examinees who had all signs of bacterial vaginosis and also isolated G. vaginalis, in 64,70% of the examinees with isolated G. vagina*lis*, 40% of the examinees with the discomforts typical

CONCLUSION

for bacterial vaginosis, and they was not found in the other examinees. According to our results, the clue cells finding is the most specific sign of bacterial vaginosis. During the last five years, the examination of vaginal smear coloured according to Gram has been increasingly recommended for determination of preliminary diagnosis of bacterial vaginosis (9,12). Examining direct microscopic preparations of vaginal discharge coloured according to Gram and ranked by Nugent scoring system in our test group of the examinees, we found the change of the various bacterial morphotypes relations in 48,80% of the examinees which tells about the presence of bacterial vaginosis. G. vaginalis was isolated in 78,04% of the examinees. Certainly, microscopic examination of vaginal smears and the interpretation of results are subjected to numerous mistakes such as: taking of materials, making and colorization of preparations, microbiologist's experience and knowledge. However, this method is very good, cheap and results can be obtained within half an hour, and comparing to cultivation it gives similar results. Almost all authors agree in the assessment that cultural examination of vaginal discharge with the purpose of determination of routine diagnosis of bacterial vaginosis is not a method of choice, because its procedure is complicate and expensive (27, 28, 29). The finding of G. vaginalis itself does not mean disease at the same time because it can be isolated in 5-60% of healthy women (30, 29). On the other side, Gardner and Dukes isolated G. vaginalis in 93% of women with the diagnosis of bacterial vaginosis (31). In our research, G. vaginalis was isolated in 63,41% of the examinees with all signs of bacterial vaginosis, in 36,59% of the examinees with one or more clinical signs of bacterial vaginosis and in 2,58% of the control group examinees without clinical signs.

Diagnosis determination for bacterial vaginosis was conducted on the basis of three out of four internationally accepted criteria according to Amsel and isolation and identification of *Gardnerella vaginalis* (*G. vaginalis*) by standard microbiological procedures. Bacterial vaginosis was diagnosed in 20,5 % women who asked for gynaecologist's help due to their personal discomfort, since significantly higher percentage of diagnosed bacterial vaginosis of 48,80% was determined in women with personal discomfort typical for this disease. Bacterial vaginosis is significantly more frequent occurrence in women who are not married, since the number of sexual partners, the time of the first sexual intercourse, the use of intrauterine contraceptive device and smoking do not cause the genesis of bacterial vaginosis. An increased vaginal discharge with unpleasant odour after sexual discourse, its pH>4,5, a positive amino odour test, an occurrence of clue cells in a direct microscopic concoction of vaginal discharge and assessment of the state of vaginal flora for bacterial vaginosis are significantly more frequent occurrences in women with individual discomforts. It was proved that *G. vaginalis* is a dominant micro organism in 95% of women with clinical signs of vaginosis although it was isolated from vaginal discharge in 40 to 50% of healthy women. In our research, *G. vaginalis* was isolated in 63,41% of examined women with all signs of bacterial vaginosis, in 36,59% of examined women with one or more clinical signs of bacterial vaginosis and in 2,58% of examined women of control group without clinical signs.

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