

Nugent Score and Vaginal Flora Abnormalities Seen in the Laboratory of University Hospital of Befelatanana

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Abstract

Background: Vaginal flora abnormalities are common in women in their genitally active period. The aims of this study are to describe vaginal flora abnormalities and genital infections discovered from the Nugent score and to describe the associated factors with these abnormalities.

Methods: This is a prospective and descriptive study of the microbiological results of the cervico-vaginal smears from the Nugent score and the microbiological culture for a period of six months from July to December 2019. This study was carried out in the laboratory of the University Hospital Joseph Raseta Befelatanana Antananarivo in Madagascar.

Results: Among of the 161 cervico-vaginal smears, the score de Nugent associated of the microbiological results showed 52 (32.3%) cases of intermediate vaginal flora, 47(29.2%) cases of bacterial vaginosis, 46 (28.6%) cases of normal vaginal flora, 13 (8.1%) cases of vaginal candidiasis and 3(1.9%) cases of *Staphylococcus aureus* vaginitis. Concerning the Nugent score values, scores 4 and 7 are the most common. Concerning the associated factors with these vaginal flora abnormalities, women over 40 years ($p=0.20$; NS), hospitalized ($p=0.25$; NS) and suffering from leucorrheas ($p=0.02$) are the most affected by these abnormalities.

Conclusion: Associated with the result of microbiological culture, the Nugent score helps biologists to diagnose genital infections from the cervico-vaginal smear.

Keywords – Nugent, cervico-vaginal smear, bacterial vaginosis.

I. INTRODUCTION

Vaginal flora abnormalities consist of disturbances of the normal vaginal flora and characterized by replacement of lactobacilli with a variety of anaerobic bacteria and mycoplasmas [1]. These disturbances can be of intermediate

degree, corresponding to the intermediate vaginal flora, or be deeper, constituting bacterial vaginosis. Currently, bacterial vaginosis is the most common female genital infection in the world [1]. However, it is more common in developing countries. Bacterial vaginosis is associated with several important health problems such as genital infections [2],

premature birth [3], spontaneous abortions [4] and in vitro fertilization failures [5]. For the diagnosis of these anomalies of the vaginal flora, there is a score called “Nugent score” which allows classifying the different types of vaginal flora. The Nugent score is a simple and reliable method for the diagnosis of bacterial vaginosis that can be adapted even in the resource poor settings [6]. The aims of this study are to describe vaginal flora abnormalities and genital infections discovered from the Nugent score during cervico-vaginal smear and to describe the associated factors with these abnormalities.

II. MATERIALS AND METHODS

This is a prospective and descriptive study of the microbiological results of the cervico-vaginal smears from the Nugent score and the microbiological culture for a period of six months from July to December 2019 in the laboratory of the University Hospital of Befelatanana. Cervico-vaginal smears were performed in hospital wards or in the laboratory.

Two sterile swabs were used to collect the endocervix and exocervix. A disposable speculum is used for each cervico-vaginal smear except for virgin girls and women who undergo a simple vulvar swab. In the laboratory, microbiological culture was done on 2 types of agar. The first

is a blood agar for the endocervix swab. It is incubated in the oven at 37°C for 48 hours in an atmosphere rich in CO₂. The second is an ordinary chromogenic agar called “Uriselect®” for the exocervix swab. It is also incubated in the oven at 37°C for 24 hours. The CO₂ is not necessary. Blood agar is necessary for the culture of bacteria species *Neisseria gonorrhoeae*. Uriselect® agar is used for the detection of vaginal candidiasis and other germs.

After the culture, the sample is spread on a slide with physiological serum covered with a coverslide followed by microscopic examination. Likewise, the sample is spread on a slide followed by staining of Gram and microscopic examination. These microscopic examinations will allow visualizing and counting the elements which are present in the vaginal flora and establish the Nugent score to be able to classify the vaginal flora of the patient. Indeed, Gram-stained vaginal smears were scored with the Nugent method and categorized as bacterial vaginosis-negative (Nugent score, 0-3), bacterial vaginosis-intermediate (Nugent score, 4-6), or bacterial vaginosis-positive (Nugent score, 7-10) [7-8]. The table 1 shows the names and quantities of the elements found, the corresponding Nugent score and the corresponding type of vaginal flora [7-8].

Table 1: Nugent score and significance

Elements found on microscopic examination	Nugent score	Vaginal flora abnormalities
- lactobacilli: very large quantity	0	Normal vaginal flora
- lactobacilli: large quantity	1	Normal vaginal flora
- lactobacilli: medium quantity	2	Normal vaginal flora
- lactobacilli: small quantity	3	Normal vaginal flora
- Lactobacilli : rare - Other bacteria (mobilincus...): small quantity	4	Intermediate vaginal flora
- Lactobacilli : rare - Other bacteria (mobilincus...): medium quantity	5	Intermediate vaginal flora
- Lactobacilli : rare - Other bacteria (mobilincus...): large quantity	6	Intermediate vaginal flora
- Disappearance of lactobacilli - Very abundant anaerobic and polymorphic flora (Gardnerella vaginalis....)	7	Bacterial vaginosis
	8	Bacterial vaginosis
	9	Bacterial vaginosis
	10	Bacterial vaginosis

The parameters studied were age, clinical information, department, score de Nugent and microbiological results. The data entry and processing was performed on the software Epi-info 3.5.2. The comparison of percentages used the Chi square tests. For ethical reasons, the authorization of the

director of establishment was obtained before the data were collected in the registers. The seizure was done anonymously to maintain confidentiality. The statistical significance threshold used was $p = 0.05$.

III. RESULTS

1.1. Microbiological results of the cervico-vaginal smears

Among of the 161 cervico-vaginal smears, the score de Nugent associated of the microbiological results showed 52

(32.3%) cases of intermediate vaginal flora, 47(29.2%) cases of bacterial vaginosis, 46 (28.6%) cases of normal vaginal flora, 13 (8.1%) cases of vaginal candidiasis and 3(1.9%) cases of *Staphylococcus aureus* vaginitis (figure 1).

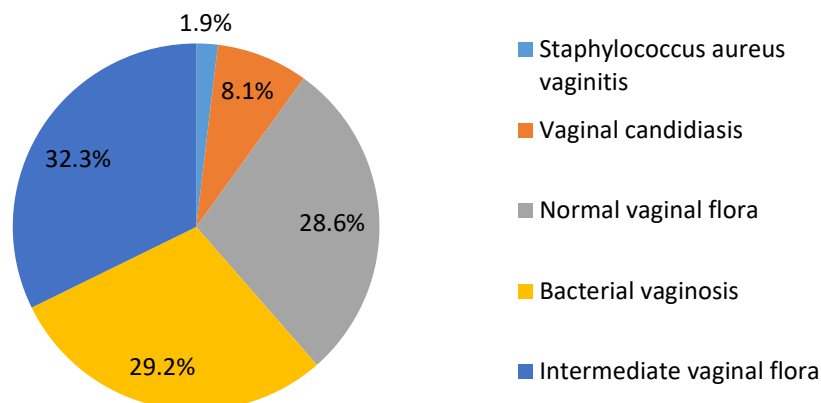


Figure 1: Microbiological results of the cervico-vaginal smears from the Nugent score and the microbiological culture

1.2. Distribution of the Nugent scores

The figure 2 shows the distribution of the Nugent scores according to the different aspects of the vaginal flora which

are represented by the normal vaginal flora (Nugent score = 0 to 3), the intermediate vaginal flora (Nugent score = 4 to 6) and bacterial vaginosis (Nugent score =7 to 10). Scores 4 and 7 are the most common.

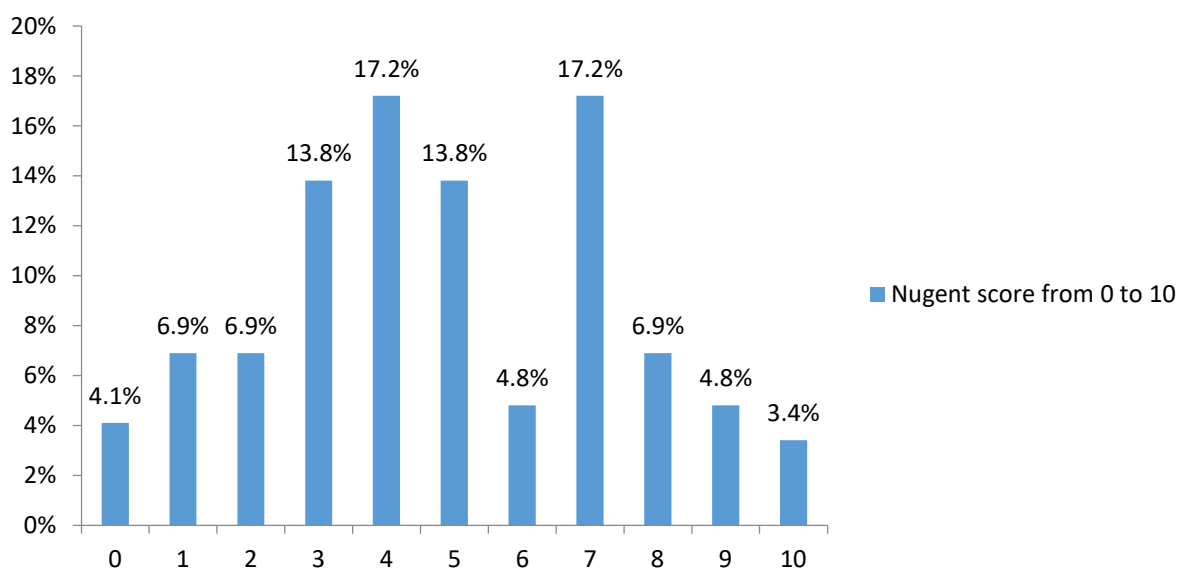


Figure 2: Nugent score

Associated factors with the vaginal flora abnormalities

Concerning the associated factors with these vaginal flora abnormalities, women over 40 years ($p=0.20$; NS), hospitalized ($p=0.25$; NS) and suffering from leucorrheas

($p=0.02$) are the most affected by these abnormalities (table 2).

Table 2: Associated factors with vaginal flora abnormalities

	Normal vaginal flora (N=46)		Vaginal flora abnormalities (N=115)		Total	p-value
	n	%	n	%		
Age (Years)						
<40	38	31.1	84	68.9	122	0.20
≥40	8	20.5	31	79.5	39	
Departments						
Outpatients	45	30.0	105	70.0	150	0.25
Hospitalized patients	1	9.1	10	90.9	11	
Total général						
Clinical information						
Others	38	33.6	75	66.4	113	0.02
Leukorrhea	8	16.7	40	83.3	48	

IV. DISCUSSION

1.3. Microbiological results of the cervico-vaginal smears and the Nugent score

Healthy vaginal flora, dominated by *Lactobacillus* species (spp.), plays an important role in the protection against genital infections [9]. Imbalances of the vaginal flora caused by a reduction in the proportion of *Lactobacillus* spp. can lead to an overgrowth of mixed anaerobic bacteria, which can ultimately result in bacterial vaginosis [10].

In this study, the microbiological results of the cervico-vaginal smears from the Nugent score and the microbiological culture showed a predominance of intermediate vaginal flora. At present, little is known about the role of the intermediate vaginal flora, which can either be regarded as the transition from a healthy to an abnormal flora or as the transition from abnormal flora back to a healthy vaginal microbiota [11]. The intermediate vaginal flora constitutes a highly heterogeneous group that may or may not include vaginal *Lactobacillus* spp. And that may or may not be characterised by the presence of anaerobic bacteria [10]. Among the intermediate vaginal flora in this study, the Nugent score 4 is the most common. This score is close to score 3 which represents the normal vaginal flora. Thus, we

can consider that the majority of women in our study have normal or slightly disturbed vaginal flora due to different situations of daily life such as taking certain drugs, taking tobacco or using soaps for personal hygiene. Thus, measures to prevent modification of the vaginal flora should be advised to women.

Concerning bacterial vaginosis, it concerns 28.6% of the women in this study. This frequency is higher compared to the study carried out in Mysore, India which found a prevalence of 19% of bacterial vaginosis among sexually active females [12]. The Nugent score 7 was the most frequent. Thus, the majority of patients have been in the first stage of bacterial vaginosis. Bacterial vaginosis is a polymicrobial syndrome characterized by loss of normal vaginal flora and acquisition of mixed anaerobic bacteria with an increase in the bacterial load from 1000 to 10,000 times above the normal [6]. Bacterial vaginosis has been described as a disease, a disorder, a vaginal inflammation, an infection, a microbial dysbiosis, a condition, and in some women, a normal situation. In order to fit the definition of a disease, bacterial vaginosis would have to be a disorder of function that produces specific signs or symptoms or affects the vagina in an aberrant way. Yet, there is little consistency in patients reporting signs and symptoms when bacterial is diagnosed,

nor the appearance of aberrations to the vagina. [13]. Even if there are no clinical signs, women with bacterial vaginosis should be treated and counseled regarding preventive measures to avoid recurrences. There are many preventive measures against bacterial vaginosis : avoid using deodorants or scented products in the vagina and on the genitals, avoid using a strong detergent to wash your underwear, change tampon or sanitary napkin frequently, wipe from front to back after using the toilet, dry the vaginal area after each shower / bath, swim, workout, or vigorous physical activity and change underwear after each swim, training or vigorous physical activity. All women who come to be consulted should be counseled regarding these preventive measures. Likewise, bacterial vaginosis should be treated properly. Indeed, without treatment, bacterial vaginosis is associated with several important health problems such as genital infections [2], premature birth [3], spontaneous abortions [4] and in vitro fertilization failures [5]. And there is a bidirectional association between bacterial vaginosis and increased risk of acquisition of sexually transmitted infections [6].

Concerning the other microbiological results in this study, they are rare but should not be underestimated. These are vaginal candidiasis and vaginitis. Vaginal candidiasis or vulvovaginal candidiasis (VVC) is a debilitating pathological condition caused by *Candida* species, commonly characterized by vulvar itching, burning, pain while urinating, and vaginal discharge [14]. *Candida* is a dimorphic fungus from the phyla Ascomycota that inhabits the respiratory, gastrointestinal and genitourinary tracts of more than 30% of healthy individuals during their lifetime [15, 16]. Alterations in the symbiotic interplay between the fungus and the mucosal ecosystem are associated with mild to moderate fungal dysbiosis, depending on the infected area as well as the patient's health status. After anaerobic bacterial vaginosis, VVC is considered the second most common vaginal infection, affecting 75–80% of women at least once in their lifetime [14]. Thus, after the laboratory diagnosis, vaginal candidiasis must be treated properly to improve the quality of life of patients.

Concerning the vaginitis, *Staphylococcus aureus* is the bacteria highlighted in this study. However, other microorganisms can be responsible for vaginitis such as enterobacteria, *Neisseria gonorrhoeae*, *Streptococcus agalactiae*, *Trichomonas vaginalis* [17]. Despite the different pathogenesis, symptoms of fungal and bacterial vaginitis are often confused, thus resulting in women having an inaccurate diagnosis and reduced quality of life.

Thus, the present study has highlighted the importance of microbiological examination of the cervico-vaginal smear and the Nugent score. They make it possible to highlight the different anomalies of the vaginal flora and other genital infections. Patients should be treated appropriately and counseled regarding preventive measures against these conditions.

1.4. Associated factors with the vaginal flora abnormalities

Concerning the socio-demographic factors, patients over 40 years old and hospitalized are the most affected by vaginal flora anomalies. Even if there is no significant difference, hospitalized patients are more exposed because of taking many drugs and the difficulty to wash in the hospital if unable to stand. Likewise, patients over 40 years of age are more affected because they also take medication in the majority of cases due to chronic diseases which generally start to appear from 40 years of age. Concerning the clinical information, leukorrhea is the most frequent with a very significant difference in this study. Leukorrhea is frequent because the cervico-vaginal smear is only analyzed in the presence of leucorrhea.

Therefore, clinicians should order a microbiological analysis of the cervico-vaginal smear if the patient has other clinical signs. Indeed, the other clinical presentations of these commons are represented by malodorous homogeneous discharge, pruritus, dyspareunia, and lower abdominal pain [6].

In brief, the cervico-vaginal smear is a very important exam and should be done in case of clinical signs suspect of genital conditions. Similarly, the Nugent score helps to highlight abnormalities in the vaginal flora.

V. CONCLUSIONS

The cervico-vaginal smear is a very important exam and should be done in case of clinical signs suspect of genital conditions. Associated with the result of microbiological culture, the Nugent score helps biologists to diagnose genital infections from the cervico-vaginal smear. The different anomalies of the vaginal flora and genital infections should be treated properly. Similarly, measures to prevent modification of the vaginal flora should be advised to women.

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