Vulvo-Vaginal Candidiasis In Pregnant Women At The Bafoussam Regional Hospital (West Region Of Cameroon) And Susceptibility Of Isolates To Antifungals

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***Abstract* - Background**: Pregnant women are more susceptible to both vaginal colonization and infection by yeast. Owing to this fact and considering the lack of data on this subject in the West Cameroon, the main objectives of this study was to determine the prevalence of vulvo-vaginal candidiasis (VVC) in pregnant women at the Bafoussam Regional Hospital and the susceptibility of isolates to some commonly used antifungal agents.

Methods: We conducted a cross-sectional study during a period of 3 months (June to August 2018) at the Regional Hospital of Bafoussam. A total of 143 of vaginal swabs collected were cultured on ChromAgar *Candida* medium for isolation and identification of the *Candida* species. The microdilution technique was used for the susceptibility testing of the isolates.

Results: Mycological examination revealed that, out of 143 vaginal swabs samples collected, 47 (32.86%) was positive to *Candida* spp culture. The identification of yeast isolates showed that *Candida albicans* was the major species (44.15%) followed by *Candida parapsilosis* (40.25%). Women in the third trimester of pregnancy were the most affected (75%) by the vulvo-vaginal candidiasis. The antifungal susceptibility of the isolates showed high resistance to ketoconazole (68.83%). However, sensitivity was also observe with fluconazole (24.67%).

Conclusion: The present study reveals a prevalence (32.86%) of vulvo-vaginal candidiasiscases among pregnant women at the Bafoussam Regional Hospital. *Candida albicans* (44.15%) being the most common specie involved.

Keywords—Vulvo-vaginal candidiasis, pregnant women, antifungal susceptibility.

BACKGROUND

Candida spp are opportunistic fungal pathogens, which can harmlessly colonize the skin, the urogenital system, mouth and the gastrointestinal tract [**1**]. However, they may become 'pathogenic' if the immune system is compromised, therefore cause infections, by attacking the skin, internal organs and mucous membranes including vaginal mucosa [**2**]. The infection of vaginal mucosa generally referred to as a vulvovaginal candidiasis (VVC). This infection is characterised by abundant vaginal discharge; vaginal itching, burning sensation, unpleasant odors, pain or irritation during intercourse or during the passage of urine [**3**]. Recurrent vulvovaginal candidiasis is a common cause of significant morbidity in women affecting millions of them worldwide [**4**]. About 25% of women experience a vulvo-vaginal candidiasis and 70 to 75% of women who have an active sex life suffer from it [**5**]. In addition, vulvo-vaginal candidiasis has a very negative impact on the quality of life of patients and induces significant health costs [**3**]. It is the most frequent reason for medical consultation in adult female population since it is believed that higher estrogen levels and higher glycogen content in vaginal secretions during pregnancy increase a woman's risk of developing vulvo-vaginal candidiasis [**6**]. Moreover, vulvo-vaginal candidiasis may cause systemic infections in neonate particularly with low birth weight and prematurity after delivery [**7**]. The lack of information on the genesis of recurrences and the poor results of long-term prescribed treatment, contribute to the very negative impact of this infection [**8**].

In Cameroon, very limited data exists on vulvovaginal candidiasis in pregnant women, especially in the West Region of Cameroon. A study conducted in northern Cameroon by Vroumsia *et al*., in 2013 showed a prevalence rate of 55.40% of vulvo-vaginal candidiasis in pregnant women with abnormal leucorrhoea [**9**]. Similar study conducted in Yaoundé by Kechia *et al.,* in 2015 showed a prevalence of 35.52% [**10**]. However, the changing in the epidemiology of vulvo-vaginal candidiasis requires an adjustment of strategies to control this infection by means of antifungals that act on *Candida* species. The growing number of clinical isolates resistant to current antifungal treatments, the lack of updated epidemiological data that could be used as a reference for prescription of antifungals and the lack of monitoring programs for vulvo-vaginal candidiasis suggest the need for a situation analysis concerning the antifungal susceptibility patterns of *Candida* species involved in these affections. It has been reported that, there are regional differences in antifungal drug susceptibility of *Candida* species, even within the same country, therefore these data are of great importance to implement the *Candida* species prevalence and susceptibility testing programmes in the countries [**11**]. Knowing that pregnancy is a predisposing factor for vulvo-vaginal candidiasis, and that the management of this infection remains a real problem, this study was undertaken to determine the prevalence of vulvo-vaginal candidiasis in pregnant women at the Bafoussam Regional Hospital, and the susceptibility of the isolates to some commonly used antifungals.

Methods

Type and duration of the study

It was a cross-sectional study, spreading over a period of three months from June to August 2018, at the Bafoussam Regional Hospital in the west region of Cameroon.

Study population

Our study population consisted of pregnant women (143), coming to the Bafoussam Regional Hospital for consultation. Pregnant women undergoing antifungal therapy or having a history of taking antifungals were excluded.

Sample

After cleansing the vaginal margins with Dakin's solution, vaginal swabbing (at the cervix margins) was performed using specula and sterile swabs. Each sample collected was preceded by a questionnaire.

Mycological examination and identification

The mycological examination consisted of culturing on the CHROMagar *Candida* medium (Media Mage, Johannesburg, South Africa), the various samples. This permitted us to isolate and identify (depending on the color) of *Candida* colonies after 24 to 48 hours of incubation. A number of colonies greater than or equal to 10 of a vaginal sample culture is considered to be a pathogenic character of the isolated yeast [**12**]. After identification, the *Candida* species were subjected to an antifungal susceptibility test.

Susceptibility of isolates to antifungals

The susceptibility of the isolates was tested against five (05) antifungals including fluconazole (FLC), nystatin (NYS), amphotericin B (AMB), ketoconazole (KTC) and terbinafine (TBF). MICs were determined by the liquid microdilution technique described by the CLSI (Clinical and Laboratory Standard Institute) protocol [**13**]. The MIC values obtained were compared with the standard threshold values proposed by Thérèse *et al.,* [**14**].

Ethical considerations

Within the framework of the respect of the ethics of the research, the study was submitted to the National Research Ethics Committee for Human Health (CNERSH) to obtain their approval and an ethical clearance was issued under the reference N° 2018/01/973/CE/CNERSH/SP. In addition, informed consent was obtained from patients who agreed to participate in the study.

Statistical analysis of the data

The data collected was analyzed statistically using SPSS software version 21.0. The EXCEL 2013 software allowed us to create the graphics. Only factors that had a P value <0.05 (Chi -Square test) were significant.

Results

Of the 143 pregnant women who agreed to participate in the study, 47 had vulvo-vaginal candidiasis, a prevalence rate of 32.86% in the study population. There was a statistically significant difference (*p*-value = 0.0006) between the prevalence of vulvo-vaginal candidiasis in relation to age (**Table 1**) and the group of women received in the third trimester of pregnancy was the most affected by vulvo-vaginal candidiasis (75%). Also, there was a significant association between the trimester of pregnancy and vulvovaginal candidiasis (p-value = 0.048) (**Table 1**). We equally found no significant difference between the prevalence of VVC in relation to marital status (P-value = 0.109) and the educational level (P-value = 0.457) (**Table 1**). The higher risk factors were found in all pregnant women who had vulvo-vaginal candidiasis. In some, many of these risk factors were found. Wearing fair / synthetic underwears was the most frequently identified risk factor (53.19%), followed by significant sweating (24.43%). All these

few considered, were significantly related to VVC (**Table 1**). The present study reveals the presence of some women with asymptomatic vulvo-vaginal candidiasis (25.53%). The rest (74.46%) had a symptomatology indicative of vulvovaginal candidiasis. Two main clinical signs were observed, vulvar pruritus (42.55%) and leucorrhoea (46.80%). The presence of each of these two clinical signs as well as dyspareunia and vulvar burns were statistically related to vulvo-vaginal candidiasis (**Table 1**). In these pregnant women, the frequency of the species was variable (Figure 1). *Candida albicans* (44.15%) and *Candida parapsilosis* (40.25%), were the most frequent species, followed by *Candida dubliniensis* (6.49%), *Candida tropicalis* (3.89%), *Candida krusei* (3.89%) and finally *Candida glabrata* (1.15%). (**Figure 1**).

Table 1: Factors related to vaginal candidiasis in pregnant women.

Factors	Description	Positive (n=47)	%	P-value
Age	[16-26[	26	55.31	0,0006
	[26-36[	20	42.55	
	[36-46[	1	2.12	
	≥46	0	0.00	
Gestational age	1st trimester	10	21.27	0,048
	2e trimester	16	34.,04	
	3e trimester	21	44.68	
Marital status	Married	29	61.70	0.109
	Not married	18	38.29	
Level of education	Primary school	8	17.02	0.457
	Secondary school	26	55.31	
	Higher education	13	27.66	
Risk factors	Wearing synthetic underwear	25	53.19	0.005
	Deep personal hygiene practice	19	40.42	0.000034
	Significant perspiration	22	46.80	0.000036
	Arse wiping	14	29.78	0.034
	Vulvar burns	5	10.63	0.039
Clinical Signs	Leucorrhoea	22	46.80	0.0000
	Vulvar pruritus	20	42.55	0.000
	Vaginal dryness	3	6.38	0.054
	Dysuria	2	4.25	0.107
	Dyspareunia	6	12.76	0.028

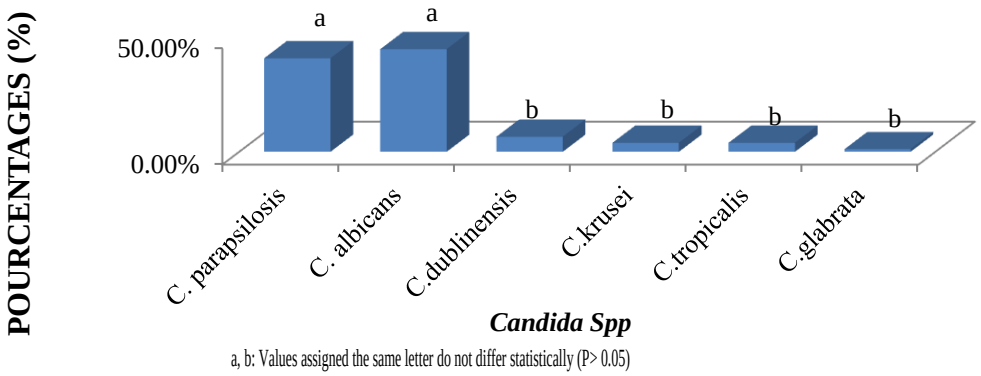

Figure 1: Distribution of different *Candida* species identified

The MIC values ranged from <0.125 to >1024µg/mL. The distribution of isolates, according to whether they were susceptible, intermediate or resistant to the antifungals used, can be seen in **Table 2**. This table shows that the isolates were more sensitive to fluconazole (4.67%), nystatin (20.77%) and amphotericin B (20.77%). In contrast, the majority of isolates were resistant to ketoconazole (68.83%) and terbinafine (66.23%).

Table 2: patterns of *Candida* isolates involved in vulvo-vaginal candidiasis to ketoconazole, fluconazole, nystatin, amphotericin B and terbinafine

Antifungals		***Ca*** **(n=34)**	***Cp*** **(n=31)**	***Cd*** ***(n=5)***	***Ct*** **(n=3)**	***Ck*** **(n= 3)**	***Cg*** **(n=1)**	**Total**
Nystatin	S	6(17.64%)	5(16.12%)	3(60%)	-	1(33.33%)	1(100%)	16(20.77%)
	I	16(47.05%)	9(29.03%)	1(20%)	1(33.33%)	2(66.66%)	-	29(37.66%)
	R	12(35.29%)	17(54.83%)	1(20%)	2(66.66%)	-	-	32(41.55%)
Amphotericin B	S	8(23.52%)	4(12.90%)	3(15%)	-	-	1(100%)	16(20.77%)
	I	14(41.17%)	14(45.16%)	1(20%)	2(66.66%)	-	-	33(42.85%)
	R	12(35.29%)	13(41.93%)	1(20%)	2(66.66%)	-	-	28(36.36%)
Terbinafine	S	0(0%)	0(0%)	0(0%)	0(0%)	0(0%)	1(100%)	1(1.29%)
	I	11(32.35%)	8(25.80%)	4(80%)	1(33.33%)	1(33.33%)	0(0%)	25(32.46%)
	R	23(67.64%)	23(74.19%)	1(20%)	2(66.66%)	2(66.66%)	0(0%)	51(66.23%)
Fluconazole	S	8(23.52%)	7(22.58%)	2(40%)	1(33.33%)	-	1(100%)	19(24.67%)
	I	4(11.76%)	4(12.90%)	-	-	2(66.66%)	-	10(12.98%)
	R	22(64.70%)	20(64.51%)	3(15%)	2(66.66%)	1(33.33%)	-	48(62.33%)
Ketoconazole	S	7(20.58%)	4(12.90%)	2(40%)	-	-	1(100%)	14(18.18%)
	I	5(17.70%)	3(9.67%)	1(20%)	-	1(33.33%)	-	9(11.68%)
	R	22(64.70%)	24(77.41%)	2(40%)	3(100%)	2(66.66%)	-	53(68.83%)

Ca*: Candida albicans,* ***Cp****: Candida parapsilosis,* ***Cd:*** *Candida dubliniensis,* ***Ct:*** *Candida tropicalis,* ***Ck****: Candida krusei,* ***Cg:*** *Candida glabrata,* **S**: susceptible, **I**: intermediate, **R:** resistant

Discussion

During the period from June to August 2017, 143 pregnant women who came for consultation to the prenatal consultation service of the Regional Hospital of Bafoussam were recruited. After mycological examination of the cervico-vaginal samples, it was found that 47 of the 143 pregnant women participating in the study had vulvo-vaginal candidiasis; a percentage of 32.86%. This frequency is almost similar to the results obtained by Okonkwo *et al*. in Nigeria in 2010 [**15**] where the studied population was of 900 pregnant women, and in Cameroon (Yaoundé) by Kechia *et al.,* [**11**] where the studied population was of 397 pregnant women. They found frequencies of 30% and 35.92% respectively. On the other hand, another study carried out in Cameroon (More precisely in the North region) by Vroumsia *et al*., [**10**] revealed a frequency of 55.40% which was higher than that of the present study. The frequency obtained by Vroumsia *et al*. would be much higher than the present study in that they focused only on pregnant women with suspicious vaginal leucorrhoea. The distribution of vulvo-vaginal candidiasis by age group among pregnant women in this study reveals that pregnant women aged 16-36 were the most affected. This observation is identical to that observed by the majority of authors who have shown that vulvo-vaginal candidiasis affects the young woman to a greater extent, and to a lesser extent, the elderly woman or the girl child [**15, 16**]. This is justified by the fact that this age group in women is characterized by intense sexual activity. It is also during this period that there is a break in the vaginal balance sometimes due to strong discharges of certain hormones (estrogen) that increase the level of glycogen (a sugar) in the vagina, which would stimulate the proliferation of *Candida* at the origin of vulvo-vaginal candidiasis [**17**]. The study of the frequency of vulvo-vaginal candidiasis according to the gestational trimester reveals in this study that women received in the third trimester of pregnancy were the most affected by vulvo-vaginal candidiasis (75%). This is in agreement with the results obtained by other authors [**18, 19, 20**]. This can be explained by the fact that the decrease in immunity that increases with gestational age is at the origin of this high frequency of vulvo-vaginal candidiasis in women in the third trimester of pregnancy [**15, 16**]. However, other authors have reported that *Candida* infections during pregnancy were quite common during the second trimester of pregnancy [**21, 22**]. The search for the presence of vulvo-vaginal candidiasis in the group of married and unmarried women showed that married women were more affected by vulvo-vaginal candidiasis (61%) than unmarried women (38.29%). This result is similar to that found by Mudeke in 2010 [**23**]. Nevertheless, even if the presence of vulvo-vaginal candidiasis in the pregnant woman is not significantly related (p-value = 0.109) to the fact that they are married or not,

it is difficult for us to exclude that fact in the VVC occurrence. The high vulvo-vaginal candidiasis rate in the married group may be explained by the fact that the frequency of pregnancy and use of contraceptives among married women is often higher than among unmarried women [**23**]. Pregnant women with a high school education were most affected by vulvo-vaginal candidiasis (55.31%), followed by those with University education (27.66%). This result corroborates that found by Kechia *et al*., in 2015 [**11**]; It could be explained by the fact that risk factors for VVC may be present in all pregnant women regardless of their level of education [**11**]. However, several authors believe that less educated women are more vulnerable to VVC [**15, 17**]. The symptoms of vulvo-vaginal candidiasis are often characteristic and occur suddenly. The intensity of symptoms varies from woman to woman and in rare cases, women may have no symptoms [**4**]. The present study allows us to note a fairly important frequency of asymptomatic vulvovaginal candidiasis in pregnant women (25.53%), a result close to the 26.89% obtained by Okonkwo *et al.* [**15**] and 29.79% obtained by Kechia *et al.,* [**11**]. This study revealed two main clinical signs, vulvar pruritus (20/47; 42.55%) and leucorrhea (22/47; 46.80%). The presence of each of these clinical signs was statistically related to vulvo-vaginal candidiasis (respective p-values of 0.000 and 0.0000). This result corroborates the results obtained by other authors [**11, 24**]. The present study also shows that vulvar burns and dyspareunia are also statistically related to vulvo-vaginal candidiasis (*p*-value of 0.039 and 0.028, respectively). Even though VVC can be contracted during sexual intercourse, the latter is not the only cause of this infection. Several factors can promote the triggering of vaginal mycosis. Only a few risk factors for vulvo-vaginal candidiasis were the subject of this study. The results of this study show that the wearing of fair underwear and significant sweating, are significantly related to vulvo-vaginal candidiasis. Its results are similar to those obtained by Kechia *et al.* [**11**]. In addition, the present study notes that wiping from the back to the front and deep intimate toilet was also significantly related to vulvo-vaginal candidiasis. The mycological examination of the various samples made it possible to identify 77 yeast strains. They were identified thanks to the specific medium CHROMagar *Candida*. Thus we have identified six (06) yeast species of the genus *Candida*. Of all identified strains of the specimens, *Candida albicans* was predominantly isolated (44.15%), followed by *Candida parapsilosis* (40.25%), *Candida dubliniensis* (6.49%), *Candida krusei* (3.89%), *Candida tropicalis* (3.89%) and finally *Candida glabrata* (1.15%). This presence of *Candida* species isolated in this study is not identical to that described by the majority of authors [**2, 11, 25**]. Although *Candida albicans* remains the most isolated species in this study, as in other studies, we still note in the present study compared to others, a difference in the frequency of isolated species as well as the absence of some other *Candida* species. The difference in the frequency of isolated species can be explained by the fact that our study population was small compared to other studies. In addition, the absence of some *Candida* species among the species isolated in the present study can be explained by the fact that the specific medium CHROMagar *Candida* only allows the presumptive identification of certain species like *Candida glabrata*, *Candida albicans*, *Candida tropicalis*, *Candida krusei*, *Candida parapsilosis* and *Candida dubliniensis* from primary clinical specimens [**26**].

Candida species have several levels of susceptibility to frequently used antifungal drugs. The results of the sensitivity test showed that the percentage of sensitivity of azoles (21.42%), was higher than that of polyenes (20.77%). In addition, the isolates were more sensitive to fluconazole (24.67%). This result corroborates that of Sobel in 2003 [**27**], which has achieved good sensitivity results with an azole antifungal agent (fluconazole). As expected, the isolates were very resistant to terbinafine (66.23%). Terbinafine is a synthetic antifungal drug with fungicidal activity against dermatophytes, molds and fungistatic activity against *Candida* species [**28**]. Unlike the results obtained by Salehei *et al*., [**29**] who achieved good results for terbinafine against *Candida* spp. several authors have reported that this drug does not show significant *in vitro* activity against *Candida* yeasts [**30**].

The overall results showed that, of the 143 pregnant women in the study, 32.86% had vulvo-vaginal candidiasis. *Candida albicans* was the predominantly isolated species (44.15%); however, it is not the only species involved in vulvo-vaginal candidiasis. Overall, the isolates identified were very resistant to the antifungals. We noted that the isolates were highly resistant to ketoconazole (68.83%). The highest percentage of sensitivity was obtained for fluconazole (24.67%).

Conclusion

Our work indicated that vulvovaginal candidiasis is relatively common in the west region of Cameroon. Several factors were found to be associated with the occurrence of vulvo-vaginal candidiasis. Therefore, in addition of identifying the causative agent and performing antifungal susceptibility test before any treatment, the effects of risk factors should be addressed in order to reduce the incidence of vulvo-vaginal candidiasis in women.

What is already know on this topic

- It's well known that pregnancy is a predisposing factor for vulvo-vaginal candidiasis, and that the management of this infection remains a real problem.

What this study adds

- This study provided data regarding the prevalence of vulvo-vaginal candidiasis in pregnant women at the Bafoussam Regional Hospital, and the antifungal susceptibility patterns of the aetiological agents.
- Data provided in this study will contribute to a better management of this infection among the affected population

Competing interests

All the authors have read and agreed to the final manuscript.

Authors' contributions

The authors declare no competing interests.

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Tables and figures

Table 1: Factors related to vaginal candidiasis in pregnant women.

Table 2: Susceptibility patterns of *Candida* isolates involved in vulvo-vaginal candidiasis to ketoconazole, fluconazole, nystatin, amphotericin B and terbinafine

Figure 1: Distribution of different *Candida* species identified.

REFERENCES

[1] A. D. Dantas, K. K. Lee, I. Raziunaite et al., "*Cell biology of Candida albicans-host interactions", Current Opinion in Microbiology*, vol. 34, pp. 111-118, 2016.

[2] A. Sellam and M. Whiteway, "Recent advances on Candida albicans biology and virulence"*, F1000Res*, vol. 5, p. 2582, 2016.

[3] M. Belayneh, M. E. Sehn, and C. Korownyk, "Recurrent vulvovaginal candidiasis*". Canadian Family Physician*, vol. 6, no. 63, p. 455, 2017.

[4] J. D. Sobel, "*Recurrent vulvovaginal candidiasis", American Journal of Obstetrics Gynecoly*, vol. 1 , no. 214, pp. 15-21, 2016.

[5] A. Matheson and D. Mazza, "Recurrent vulvovaginal candidiasis*:* A review of guideline recommendations"*, Australian & New Zealand Journal of Obstetrics & Gynaecology*, vol. 2, no. 57, pp. 139-145, 2017.

[6] B. Goncalves, C. Ferreira, C. T. Alves et al., "Vulvovaginal candidiasis: Epidemiology, microbiology and risk factors*", Critical Reviews in Microbiology*, vol. 6, no. 42, pp. 905-927, 2016.

[7] L. Filippi, C. Poggi, E. Gozzini et al., ""Neonatal liver abscesses due to Candida infection effectively treated with caspofungin"*, Acta Paediatrica*, vol. 5, no. 98, pp. 906-909, 2009.

[8] M. Lachowsky and D. Winaver, "*Avant-propos. Aspects psychosomatiques de la consultation en gynécologie",* Elsevier*,* Paris, 2007. Doi : 10.1016/B978-2-294-06816-4.50021-1.

[9] T. Vroumsia, D. Moussa, G. Bouba, et al., "Prevalence of Vulvovaginal Candidiasis amongst pregnant women in Maroua (Cameroon) and the sensitivity of Candida albicans to extracts of six locally used antifungal plants"*. International Research Journal of Microbiology*, vol. 3, no. 4, p. 89-97, 2013.

[10] F. A. Kechia, J. S. Dohbit, E. A. Kouotou et al., **«**Profil Épidémiologique et Étiologique de la Candidose Vulvo-Vaginale chez la Femme Enceinte à Yaoundé (Cameroun)", *Journal of medicine and health Sciences,* vol. 4, no. 16 , pp. 1-5, 2015.

[11] C. W. Africa, and P. M. Abrantes, "Candida antifungal drug resistance in sub-Saharan African populations: A systematic review version 2*", F1000Res*, vol 5, p. 2832, 2016.

[12] J. P. **Bouchara, M. Pihet, L. Gentile et al., "**Les levures et les levuroses, Cahiers de formation Biologie médicale", *imprimerie vert, Paris France*, no. 44, 200p, 2010.

[13] CLSI (Clinical and Laboratory Standard Institute), "Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts. *Approved standard, M27-A3"* Wayne, P A, 2008*.*

[14] K. L. Therese, R. Bagyalakshmi, H. N. Madhavanet et al., "*I*n-vitro susceptibility testing by agar dilution method to determine the minimum inhibitory concentrations of amphotericin B, fluconazole and ketoconazole

against ocular fungal isolates", I*ndian Journal of Medecinal and Microbiology*, vol. 4, no. 24, pp. 273-279, 2006.

[15] N. J. Okonkwo and P. U. Umeanaeto, "Prevalence of Vaginal Candidiasis among Pregnant Women in Nnewi Town of Anambra State", *African Research Review*, vol. 4, no 4, p. 539-548, 2010.

[16] M. B. Seema, M. P. Rajaram, A. P. Sunanda et al., "Prevalence of non-albicans Candida infection in Maharashtrian women with leucorrhea". *Annals of Tropical Medicine and Public Health*, vol. 2, no. 5, pp. 119-123, 2012.

[17] D. J. Xu and J. D. Sobel, "Candida Vulvovaginitis in Pregnancy. Division of infectious diseases" , *Current Infectious Diseases Report*, vol. 6, no. 6, pp. 445-449, 2004.

[18] B. Balaka, A. Agbéré, A. Dagnara, et al., "Genital bacterial carriage during the last trimester of pregnancy and early-onset neonatal sepsis", *Archives de Pediatrie*, vol. 5, no. 12, pp. 514-519, 2005.

[19] T. G. Bauters, M. A. Dhont, M. I. Temmerman et al., "Prevalence of vulvovaginal candidiasis and susceptibility to fluconazole in women", *American Journal of Obstetrics and Gynecoly,* vol. 3, no. 187, pp. 569-574, 2002.

[20] L. M. Linhares et al., "Differenciation between women with vulvovaginale symptoms who are positive or negative for *Candida* species by culture", *Infection Diseases and Obstetrics Gynecoly*, vol. 4, no. 9, pp. 221-225, 2001.

[21] J. Robert. «Infections de levure pendant la grossesse». http://www.doctissimo.fr/grossesse/diaporamas/les-infections-vaginales-pendant-la-grossesse, 2017 17/08/2017, [cited 2019 17 October].

[22] H. Jamilih, Les Candidoses vulvo-vaginales chez la consultante à l'Hôpital militaire d'instruction Mohamed V de Rabat : étude prospective 2009-2010. Thèse de Doctorat en Pharmacie. Université Mohammed V, Rabat-Maroc. 2010.

[23] M. H. Mudeke, " Etude épidémiologique sur les vulvo-vaginites à *Candida albicans* chez les femmes adultes dans la ville de Kinshasa. Cas de l'Hôpital Saint-Joseph. Thèse de Doctorat en Médecine. Université technologique Bel Campus, Kinshasa-RDC. 2010.

[24] S. Nouraei, A. A. S. Amir, M. Jorjani, et al., "Comparison between Fluconazole with Oral Protexin Combination and Fluconazole in the Treatment of Vulvovaginal Candidiasis", *ISRN Obstetrics and Gynecoly*, doi: 10.5402/2012/375806.

[25] E. J. Baron, G. H. Cassell, L. B. Duffy et al., "Laboratory diagnosis of female genital tract infections. *Cumitech*. 1983, Washington, D.C, *American Society for Microbiology*, vol. 17, pp. 1-28, 1993.

[26] F. C. Odds and R. Bernaerts "CHROMagar Candida, a new differential isolation medium for presumptive identification of clinically important Candida species". *Journal of Clinical Microbiology*, vol. *8*, no. 32, pp. 1923-1929, 1994.

[27] J. D. Sobel, "Management of patients with recurrent vulvovaginal candidiasis", *Drugs*, vol. 11, no. 63, p. 1056-1059, 2003.

[28] C. Gianni, "Update on antifungal therapy with terbinafine", *Italian journal of dermatology and venereology* , vol. 3, no 145, pp. 415-424, 2010.

[29] Z. Salehei, Z. Seifi, A. Z. Mahmoudabadi, "Sensitivity of vaginal isolates of Candida to eight antifungal drugs isolated from Ahvaz, Iran", *Jundishapur Journal of Microbiology*, vol. 4, no. 5, p. 574-577, 2012.

[30] N. S. Ryder and B. Favre,"Antifungal activity and mechanism of action of terbinafine "*pharmacotherapy*, vol. 8, pp. 275-287, 1997.