PREVALENCE OF CANDIDIASIS AND CYTOLOGICAL CHANGES AMONG FEMALE ATTENDING FEDERAL MEDICAL CENTRE KEFFI, NASARAWA STATE, NIGERIA.

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YEPWI JULIANA

NSU/PGD/MCB/0048/18/19

POSTGRADUATE DIPLOMA IN MICROBIOLOGY.

DECEMBER, 2019

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POSTGRADUATE DIPLOMA IN MICROBIOLOGY.

A DESSARTATION SUBMITTED TO THE SCHOOL OF POSTGRADUATE STUDIES, NASARAWA STATE UNIVERSITY KEFFI, IN PARTIAL FULFILMENT OF THE REQUIREMENT OF THE AWARD OF POSTGRADUATE DIPLOMA IN MICROBIOLOGY.

DEPARTMENT OF MICROBIOLOGY
FACULTY OF NATURAL AND APPLIED SCIENCES
NASARAWA STATE UNIVERSITY,KEFFI
NIGERIA

DECLARATION.

I hereby declare that this project has been written by me and it is a report of my research work. It has not been presented in any previous application for state diploma. All quotations are indicated and sources of information specifically acknowledge by means of references.

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YEPWI JULIANA

NSU/PGD/MCB/0048/18/19

CERTIFICATION

This is to certify that this project titled "Prevalence of Candidiasis and Cytological Changes among Females Attending Federal Medical Centre Keffi, Nasarawa State, Nigeria" meets the regulations governing the award of Postgraduate Diploma in Microbiology, of the School of Postgraduate Studies, Nasarawa State University, Keffi and is approved for its contribution to knowledge.

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DEDICATION

To the Almighty God my lord Jesus Christ who was in the boat when the storm lasted for a while and the night seems so long.

ACKNOWLEDEMENT

Adoration, honour and power to my Lord Jesus Christ who is the source of my strength and inspiration, whose power in me has done infinitely more than I have asked for or could have imagined, and whose without his help I can do nothing.To you be glory and dominion both now and forever more. Amen.

Thanks to my parents Chief and Mrs. Dauda Yepwi and my special daddy Prof. Gideon Amuga who have supported me morally through this journey and my brothers Idako,Narai and Aligaza.

I sincerely appreciate my supervisor M.P.Adoga and my lecturals Dr Istifanus, Dr Ishaku Akyla, Dr Ishaleku, Prof. Obiekezie,Dr Owuna.

I want to appreciate my PGD 2018/2019 set,specially my close pals Cynthia Luku,Amos,Ruggaya Buba,Jet, Mrs Ajibose

TABLE OF CONTENT

CHAPTER THREE

MATERIALS AND METHODS

CHAPTER FOUR

RESULT

CHAPTER FIVE

DISCUSSION RECOMMENDATION AND CONCLUSION

ABSTRACT

This study was carried out to establish the prevalence of Candida infection among women in the study area, the aim of this study is to determine the prevalence of candidiasis and cytological features among female patients attending Federal the Medical Center Keffi. To find out the cytological changes of the vaginal and cervix infected with *Candida species* and also find out the predisposing factors. Four hundred (400) cervical smear samples were collected from women between of 21 and 60 attending a cervical screening exercise at Federal Medical Centre Keffi (FMCK), from January 2018 to December 2019. Some vital information was obtained from them in the form of medical history. Among the women studied, 8 out 400(2%) had Candida infection (Candidiasis). Out of the 8 samples positive for Candida, 8 (100%) showed the most predominant cytological changes which are-variability in the cell nuclei sizes, classic cell clumping and polymorphonuclear leucocyte at the smear background.6 (75%) showed a reactive intra-epithelial infiltration with leucocytes, 5 (62.5%) had slight hyperkeratosis. Those women within the age of 31-40 had more cases of candidiasis. It was found that those who were married (382) are more prone to the infection (1.75%) than the single and widows. Recurrent vaginal discharge was found to be suggestive of candidiasis as 263 women with the history had 6 (1.5%) of the total prevalence (2.0%). 1.25 of those women with the infection was found to have used one contraceptive or the other.

CHAPTER ONE

INTRODUCTION

1.1 Background of the Study

Candidiasis is a depending pathogenic fungal infection caused by yeast of the genus Candida. The genus Candida comprises of about 154 species. 20 of the species cause infection in humans. Candida is a thin-walled yeast, relatively small, forming spores that form spores. Candidiasis colonies can grow rapidly within 3 days (Candida Albans*et al.*, 2015) (CDCP *et al.*, 2014).*Candida species* (yeast) occasionally or habitually live on the skin and mucous membranes without causing harm to the host. Symptoms occur only when excessive growth of this organism occurs. Symptoms of candidiasis depend on the location of the mucosal lesion. There are three main types of candidiasis in candidiasis (Centers for Disease Control and Prevention, 2014).

Candidiasis-Oral pharynx Yeast infection of the Genital / Vulvovaginal candidiasis Invasive candidiasis-type of candidiasis that enters the bloodstream and spread throughout the body. Genital candidiasis is relatively common in the general population. About 75% of women have a yeast infection at least once in their lives (Centers for Disease Control and Prevention et al., 2014).Candida can increase and cause infection if the surrounding if the vagina changes in ways that support the growth. This is caused by hormones, drugs or changes in the immune system (Sobel, 2007). Genital candidiasis penetrates the vaginal mucous membrane and triggers an inflammatory reaction. Permanent inflammatory cells are usually polymorphonuclear cells and macrophages (Swidsinski *et al.*, 2019), budding yeast, hyphae, or pseudophytes that show the presence of candida under a microscope (Denders *et al.*, 2017).

The presence of this fungus in the vaginal vulva usually causes itching, redness of swelling (inflammation) and cracks in the vaginal wall (Farhan *et al.*,·2018), resulting in very uncomfortable white quarks. As a result of the relapse of genital candidiasis, he feels more stress due to severe clinical depression, lack of satisfaction in life, lack of self-esteem, serious mixing of sexual and emotional relations (Goncalves *et al.*, 2016).

Although candidiasis as a primary and secondary infection is caused mostly by *C. albicans*, all candida species may be mixed together in the tissue or organ and the can grow best on warm moist surface (National Institute of Allergy and Infectious Diseases,2010)thus speculation of yeast-like isolates is very important on the identification of any pathogens of vagina yeast isolated in the laboratory (CDC 2014).

The mere presence of yeast-like organism in a vagina smear does not necessarily diagnose Candida infection alone, some cytological changes of the vaginal environment had to be established before concluding the diagnosis.

Normal cervical smear has squamous epithelial cells, columnar epithelial cells, metablastic epithelial cells (transformation zone).The squamous epithelial cells contain layers of cells; superficial, intermediate, basal and parabasal cells.

Vaginal epithelial cells of fecund women who were pregnant and diabetic had a greater propensity to bind *C. albicans* than oral contraceptive user and non-diabetic controls. The highest level of adherence occurred in pregnant diabetic women. Among non-diabeticpostmenopausal females, *C. albican* is lower than for postmenopausal women diabetic women.The hormonal status of fecund and post-menopausal women assayed cytological the karyopyknotic and main ration indices, which determine the ratio of superficial, intermediate and parabasal vaginal epithelial cells. *C. albicans* adherence

increase in statue where there is an increase in number of intermediate epithelial cells. (Goncalves,2016).

Since genital candidiasis gives rise to vaginal irritation and discharge which leads to discomfort in human,there is need for precise treatment of the infection.

1.2 Statement of problem.

In developing countries including Nigeria,prevalence and cytological changes of candidiasis is unavailable and some does not exist,hence prevalence data are not available for the cytological changes.

Genital candidiasis is responsible for one third of all cases of vulvovaginitis of about 75% of child bearings women experience vaginal candidiasis at some point in their lives. About 8% of women suffer from recurrent genital candidiasis, *Candida albican* is the most causative agent (in about 90% of cases) (Center for Disease Control and Prevention, *et al.*, 2014).

1.3 Justification

To enlighten the public on the dangers of candidiasis to women health and to educate them to improve personal hygiene in order to eliminate the fungal.

To suggest that all the women that are sent to the cytology laboratories for cervical screening should also have a swab collected for specific microbial investigation.

1.4 Aim

To determine the prevalence of candidiasis and cytological features among female patients presenting at Federal the Medical Center Keffi.

1.5 Objectives

i. This study is to determine the prevalence of this infection amongst women.

ii. To study the cytological changes of the vaginal and cervix infected with candida species

iii. To identify the predisposing factor in the study area.

CHAPTER TWO

LITERATURE REVIEW

2.1 Historical background of candida

One of the principle factors of rivalry when defining thrush became whether or not it originated from the host or was an infectious agent, or an aggregate of the 2. The earliest reviews of thrush predated the concept of a microbial pathogen of the Epidemics, Hippocrates defined oral candidiasis as mouths affected with aphthous ulcerations (Dolin,2010) and this was first sketched by J.S. Wilikinsin in1849(Lynch,1994).While in 1665, Pepys Diary said 'a patient hath a fever, a thrush and a hiccup', perpetuating the idea that oral thrush originates from the host(Martins and(Jones,1940).

A Mycologists Castellani who believes in the early 1900s quoted preceding debts of thrush as 'morbid secretions of the oral mucosa'(Carderone,2002). In 1771, Rosen von Rosenstein described an invasive shape of thrush (Carderone, 2002).

In 1839, Langenbeck became credited with first recognizing a fungus in affected person with typhoid fever. Oropharyngeal and esophageal thrush with pseudo membranes have been found at post-mortem.

'Under the microscope magnified, the pseudomembranes consisted of a massive quantity of fungi'. He describes in detail what's now called septate hyphae, branched pseudohyphae and blastoconidia However, he ascribed the entity to the typhoid bacterium instead of the fungus (Knoke, 2006). In1844, J.H. Bennett found a comparable fungus within the sputum and the lungs of a patient with a pneumothorax and criticized the conclusion by using Lagenbach. The morphologic description of Bennett was essentially that as defined via Langenbeck (Carderone, 2002).

Bennett in 1844, concluded that the sickness turned into 'indicative of notable melancholy of the crucial powers and impairment of the nutritive functions of the economic system'. Once the etiology turned into conclusively tested by way of mycologists, the next factor of rivalry became the identification of the pathogen. While Langenbeck in1839, first documented the fungus related to thrush, he did not make the direct connection.

In 1923 Christian Berhourt reclassify the organism under the genus Candida which was derived from a Latin word toga candida which was a white robe that was wormed by a Roman senator (Bernett,2004). Nearly 200 species of organism are found in the genus candida which are anamorphic yeast-like cells belonging to the form- class Blastomycetes, six species,*C.albicans,C.glabrata,C.tropicalis,C.parapsilosis,C.krusei* and *C. lusitaniae* are the most commonly associated with human infection(Geogiev,2003).

2.2 Epidemiology of genital candidiasis

It is assessed that 20% of ladies might be asymptomatically colonized by vaginal yeast, (Sabel, 2007). A global prevalence of recurrent and chronic vaginal(vulvovaginal) candidiasis was carried out using UN world data population prospect of 2012 of women that have had four or more episodes of confirmed vaginal candidiasis within the period of one year.The report was across women attending gynecological clinics in China, India, USA ,Indonesia, Brazil ,Pakistan ,Nigeria, Russian Federation, Bangladesh and Japan, telephone survey (2000 women from USA) and online survey(USA, France, Italy, Spain, Germany and UK with 9% prevalence) (Corsello *et al.*, 200;Abellea *et al.*,2013) cutting across 15-64years aged female patients with 6% prevalence rate of a global burden of 134,988,103.(Chapple,2001; Mahmoudi *et al.*, 2011; UN,2012) completed the internet-based questionnaire which the majority of

them having recurrent vaginal candidiasis which are antibiotic use ranged highest (Junko *et al.*,2019).

A total of 1,217 participates, vaginal swabs were collated from the women in Greece. 75.6% of *Candida albican* 13.6% *C. glabrata* where isolated(Sofia et al,2019).

A total of 347 reproductive non-pregnant women cross-section study was carried out and 37% was prevalent with vaginal candidiasis in Sana'a city, Yemen. (Maha *et al*,2019).Total of 284 non-pregnant women(78% Caucasian,14% African American,8% Asian) In the united states there are roughly 1.4 million specialist office visits each year for candidiasis(Benedict,2019). 75% of American women in the reproductive age group harbor the organism as an opportunistic saprophyte and nearly three quarters will suffer at least one attack of the Candida vaginitis at one time or another in their lives. About the 75% of those infected will experience more than one episode, and some victims of frequent relapse and re-occurrence over a period of many years (Benedict,2019).

In western Africa, a study was conducted in middle Ghana on prevalence of vulvovaginal candidiasis among pregnant women attending antenatal clinic with prevalence of 56.4% vaginal candidiasis in 2015 of 589 pregnant women(Dennis *et al.*,2019). In the eastern part of Nigeria, the prevalence rate of 17.7% of positive vaginal candidiasis among 209 symptomatic and laboratory test in 2018 (Aniebue *et al.*, 2018) presenting at the gynecology clinic in University of Nigeria Teaching Hospital Enugu. 100 specimens tested, the overall prevalence of Candida species was 57% Research original article Azike *et al.*, 2019 (n = 57). There were 47 symptomatic and asymptomatic participants 53 participants in the study. Of the 47 symptomatic women, 36 had Candida species, while 21 of the 53 women had asymptomatic Candida species. This study found a higher prevalence of vulvovaginal candidiasis among

pregnant women that are symptomatic and asymptomatic at university hospital in Abia State. Regular checks and good hygiene practices are criticized to prevent avoidable complications.

Southern Nigeria, samples were collected from 200 female students between the age of 17-31 years from University of Calabar, 20% (40/200) were positive for Candida albican. (Mbim *et al*, 2017). North-west Nigeria , there is a rising trend in the cases of candidiasis due to its environment and uncontrolled use of very potent antibiotic.84% of the vaginal candidiasis was compared to 25% (Kwawukume and Arhin, 2002), 24% (Mirza, et al,1983),18.5%(Otero et al.,1998) and 61% (Nwadioha et al.,2013)gotten from the previous report,and this was similar to the report of Onifade and Olurunfemi in Ondo state with prevalence of 81.5%. (Onifade and Olurunfemi,2005).84% was reported as prevalence of candidiasis as at 2015 among patiences attending Aminu Kanu Teaching hospital(Ugwu,2015).

North-east Nigeria, 288 pregnant women samples were screened and 175(60.8%) were positive for candidiasis (Daniel and Swati, 2017) and was reported by Daniel and Swati, 2017 as prevalence of vaginal candidiasis in 2017 among pregnant women attending a tertiary institution(Usman Danfodiya Teaching hospital). In Niger State, total number of 40 pregnant and non-pregnant vaginal swab was collected, women attending at both General hospital Minna, Niger state resulted with prevalence rate of 47.5%(19) (Mumuney and Abalaka,2019).

In Kaduna 2017, 411 subjects participated in the study, and 66%(271) were positive. Vagina candidiasis is responsible for vaginal discharge especially child bearing age and pregnant women and is a common disease among weak immune system (Monif *et al*., 2003). (Umar, 2017).

Middle belt Nigeria, prevalence rate of 6.5% of among 200 non pregnant women attending tertiary health care facility in Abuja. (Anthony *et al.*, 2015) Prevalence of vaginal candidiasis was carried out among female students in Bingham University, Nasarawa state, in 2015 by Maikenti *et al.*, (2015).96 high vaginal swaps were collected from the female students, screened and 25(26.0%) students were infected.

In North central Nigeria, four states where selected, and the research was carried out in those states selected. This research was conducted in Federal University lafia, in microbiology department between January and to November, 2018 with 1600 samples of vaginal swabs of participant resulting to 710 (44.4%) with vaginal candidiasis (Aleruchi *et al.*, 2019).

The predisposing factors that allow the transformation from passive to active pathogen are so common, that the organism should be eradicated whenever it is found; whether the patient is symptomatic or asymptomatic because of these high incidence and prevalence figures, that candida rank second to Gardnerella /anaerobic vaginosis in frequency as a cause of vaginal distress.

2.3 Physiopathogenesis of species of candida and medical importance.

2.3.1 *Candida albican*

Candida albicans is a yeast opportunistic pathogens which is a common member of the human intestinal flora. It can also survive outside the human body (Odds, 1988). It is detected in the gastrointestinal tract and mouth in 40-60% of healthy adults (Erdogan and Rao, 2015).It is usually a commensal organism, but can be a pathogen in immunocompromised individuals under various conditions (Martins, 2014).This is one of several species of the genus Candida that cause candidiasis infections in humans, which results from an overgrowth of fungus (Erdogan and Rao, 2015).Candidiasis is

for example often observed in patients infected with HIV. *C. albicans, C. tropicalis, C. parapsilosis*, and *C. glabrata* jointly responsible for 50-90% of all cases of candidiasis in humans(Martins et al,2015).A mortality rate of 40% have been reported for patients with systemic candidiasis due to *C. albicans*(Singh et al.,2017).

C. albicans is commonly used as a model organism for pathogenic fungi (Kabir *et al.*, 2012).It is commonly referred to as a dimorphic fungus as it grows both as yeast cells and filaments. However, it has several different morphological phenotypes (Kadosh, 2019). *C. albicans* was for a long time considered as an obligate diploid organism without haploid stage. However, this is not the case. Next to the haploid phase of *C. albicans* can also exist in the tetraploid stage. The latter was formed when diploid cells of *C. albicans* mating when they are in opaque (Hickman *et al.*, 2019).The size of the diploid genome is about 29 Mb, and up to 70% of the protein coding genes have not been characterized (Sevilla and Odds,1986).

2.3.2 *Candida glabrata*

Candida glabrata is haploid yeast species of the genus Candida, formerly known as Torulopsis glabrata.No fact of sexual life cycle has been documented for this species, *C. glabrata* strains of the both mating types are usually found (Fairhead *et al.*,2008) *C. glabrata* is a commensal of the human mucosal tissue, but in the present era of broader human immunodeficiency of different causes (eg, immunomodulation therapy, longer survival with various comorbidities such as diabetes and HIV infection), *C. glabrata* is often responsible for second or third most common form of candidiasis as opportunistic organism (Fidel *et al.*,1999).*C. glabrata*infections can affect the urogenital tract or even spread infection by entering the yeast cells in the blood stream (candidemia),(Fidel *et al.*,1999)

2.3.3 *Candida tropicalis*

Candida tropicalis is a species in the genus Candida yeast. This is a common pathogen in neutropenic hosts, on the side that spreads through the bloodstream to peripheral organs (Mastromarino *et al.*,2013). For invasive disease, including the treatment of amphotericin B, echinocandins, or extended-spectrum triazole antifungal (Chai *et al*,2010)*C. tropicalis* reproduces asexually by the production of blastoconidia through budding. As blastoconidia increase in number they may elongate in shape producing structures called the pseudohyphae (Murray *et al.*,2003). Under specific conditions of reduced oxygen level in host tissues, submerged colonies in agar medium, or in the presence of 5-10% CO_2, true, septate hyphae may form.

2.4 Genital candidiasis

Vaginal candidiasis is a fungal or yeast infection in women is a lower genital tract, vulva, and vagina infection caused by *Candida spp.* (Sobel,2007). It can be termed as candidiasis or moniliasis. Vaginal candidiasis can be repeated or recurrent or relapsed relapsing (Marrazzo, 2002).Vaginal candidiasis occurs when a woman presents with four or more episodes per year. Normal vaginal characterized by dynamic interrelationship between Lactobacillus acidophilus and other endogenous estrogen, glycogen, vaginal pH, and metabolic by-product of this microbiomes. L. acidophilus produces hydrogen peroxide (as a byproduct of metabolism), which is toxic to pathogens and maintain healthy vaginal pH acidic. Vaginitis occurs when the vaginal microflora has been changed by a pathogen or biochemical changes in the vaginal environment changes environment(Oddos,1988).This biochemical changes in the vaginal environment encourage Candida population, improve their adherence to vaginal epithelial cells, and facilitate germination daughter yeast cells (Sobel,1998). These changes may alter become asymptomatic colonization of Candida infection

symptoms. Vaginal candidiasis, like many vulvar disease, has the potential to cause great psychological distress and negatively impact the quality of life of patients.

The prevalence of Candidiasis is more in pregnant women than non-pregnant women. This is characterized by low-grade inflammation of the vaginal manifesting white and yellow patches, with milky discharge on the mucosa. This infection may spread to the upper part of the thighs and vulva which cause a genital privities leading to scar tissue removal (Rhoads *et al.*, 1987).

2.5 Predisposing factors:

These are host factors that affect vaginal secretions and,thus,vaginal environment appears to play key role in causation of this disease.

2.5.1 Pregnancy:

This is the most common predisposing factor with the incidence and severity of infection increasing with the duration of gestation (Akeel *et al.*,2013;Ahmad and Khad,2009).World-wide studies show that approximately 1/3 of all pregnant women yield candida on any particular day. The high hormone levels in pregnancy and increased glycogen content of the vagina constitute a favorable environment for candida growth (Babic and Hukic,2010).At the same time, the acidity of the vaginal fluid further suppresses the growth of other microorganisms that are naturally inhibitory to candida, remaining only symbiotic lactobacilli. Pregnandiol isa steroid that is produce in large quantities during pregnancy, enhances the growth of *C. albicans* invitro. It is also possible that reduced glucose tolerance and consequent glycosuria in some pregnant women could further increase their susceptibility.

`After delivery, the precipitous drop in estrogen and progesterone levels is followed by radical changes in the vaginal micro-environment. In most patients, there is a

concomitant rapid disappearance of the clinical signs of candidiasis(Ahmad and Khan,2009).

2.5.2 Contraceptives

Some studies have shown increased Vaginal candidiasis after use of oral contraceptives with a high estrogen content (Raad *et al.*, 2003; Mohmoudi *et al.*, 2011). Increased risk of repeated infection with vaginal contraceptive sponges, diaphragms, intrauterine contraceptive devices, and condoms (Richter et al., 2005 ;. Ahmad and Khan, 2009; Grigoriou *et al.*, 2006). This is because the increase in estrogen levels that occurs with oral contraceptives increased colonization of Candida spp. And in the vaginal (Ahmad and Khan 2009; Tarry *et al.*, 2005;Consolaro *et al*, 2004;Yusuf *et al*, 2007). Al-Akeel *et al.* (2013) reported that oral-contraceptive users are more likely to develop Vaginal candidiasis compared to non-users (Al-akeel *et al.*,2013).

2.5.3 Uncontrolled diabetes mellitus

vaginal inflammation or infection, especially Vaginal candidiasis is more alarming in severe hyperglycemic conditions. Many studies show that the level of yeast colonization occurs more often in women with diabetes than non-diabetics (Ahmad and Khan, 2009; Malazy *et al*, 2007). Women with type 2 diabetes are more susceptible to colonization by *C. glabrata* (Donlan and Costerton, 2002). Farajii *et al.*, (2012) reported that Vaginal candidiasis in women with diabetes are more common than non-diabetics, and *C. albicans* is a yeast isolated the most dominant (Faraji *et al.*, 2012). Grigoriou *et al.*, (2006) reported that women with diabetes are more likely to develop fungal vaginitis compared to non-diabetics (Grigoriou *et al.*. 2006). Glucose

can stimulate the development of yeast and even encourage a change to more malignant stage.

2.5.4 Antibiotics:

Antimicrobials are thought to act as a predisposing factor by reducing the number of protective resident bacteria. The broad spectrum agents, e.g. tetracycline's, cephalosporin's and ampicillin like compounds, are the most common offenders. Adolescents or young adults who develop acne and are treated with tetracycline are prime candidates for candida vaginitis, as are women who are treated with the same drug for a chlamydial infection. After antibiotic administration, the increased prevalence of vaginal yeast colonization is usually accompanied by a simultaneous increase in the number of candida obtained from the gastrointestinal tract (Mcfarland, 2008).

2.5.5 Sexual Transmission

Due to the ubiquitous nature of candida and the innocence with which most genital infections seem to be acquired, candida vaginitis has ever been considered a sexually transmitted disease. Many have pointed to the low frequency of positive penile cultures among consort of affected women, and *Candida balantis* has been generally confined to diabetic/immunosuppressed men.

Horowitz *et al* (1987) shed more light on this concept. He cultured specimen from the vagina, rectum and mouth of 33 women with more than three culture-proven candida episodes occurring in a single year. In addition, he cultured sample from the mouth, ejaculation and rectum and their consort and found a high correction of positive cultures with the same candida species found in both partners. The oral cavities of the couple were the sites most often colonized, followed by the rectum. Although there is a

low carriage rate on penile skin and none in prostatic fluid, the 15% prevalence ejaculation colonization suggests that seminal vesicle act as an additional reservoir.

2.5.6 Feminine Hygiene practices (Douching):

Certain feminine practices such as vaginal douching have been strongly associated with BV (Holzman *et al.*, 2001;Ness *et al.*, 2002). In pregnant women, a study found significant correlation of BV with douching three or more times before or during pregnancy (Trabert&Misra,2007).

woman's timing and/or frequency of douching is highly predictive of BV (Schwebke & Weiss, 2001). However, the reason for douching by many women is because of abnormal vaginal symptoms (Ness *et al.*, 2002). It is possible BV is not the consequence of douching but rather the reason why women douche. There are however a few studies that did not find any relationship between BV and douching (Bukusi *et al.*, 2006; Demba *et al.*, 2005).

2.6 Clinical presentation

2.6.1 Signs and symptoms:

Vulva pruritis is the main symptom of candidiasis and is more intense than in the average case of trichomonomiasis. While many patients complained of vaginal itching, this probably represents vestibular pruritus since the vestibule is abundantly supplied with free nerve endings that are believed to mediate perceptions of both itching and pain. Itching varies from slight to intolerance and may interfere with normal activities and rest.

Some women only notice it prior to mense, while others complain of itching and burning immediately after intercourse. The frequency of pruritus in patients regarded

as having clinical disease based on clinical signs is about 90%. Occasionally, patients have thrush or pseudomembranous patches on the vaginal without pruritus.

Burning is also a common complain especially on urination, mostly experienced by patients who have excoriations from scratching; called vulvar dysuria. Some patients develop reflexogenic urinary urgency and frequency. Combination of dysuria and frequency can be attributed to cystitis.

Dyspareunia can be severing, particularly in nulliparas, and may progress to a tolerance of intercourse. Such women should be suspected to having vulvar vestibulitis syndrome. Speculum examination of the vagina reveals variable amounts of thick, white, curd-like or flocculent discharge which is loosely adherent to the vaginal mucosa, the discharge often likened to cottage cheese.

The mucosa beneath adherent thrust patches are usually erythematous. Upon their removal incomplete superficial ulcerations with oozing blood are occasionally seen. The acidity of vaginal secretions in candidiasis is usually within the range pH 4.0-4.7 with a Ph 4.5 mostly common. Normal secretions have pH 3.8-4.2.

2.6.1.1 Histopathology:

The changes induced in the vaginal tissue by acute candidiasis are similar to those found in acute Trichomosomiasis,although less pronounced. Intracellular oedema and hyperemia of the lamina propria are present.A chronic inflammatory infiltrate consisting primarily of lymphocytes,a few plasma cells,and occasionally,neutrophil involves the superficial lamina propria.The surface of the epithelium may exhibit debris consisting of degenerated squamous cells,neutrophils and frequently,conidia and

hyphae of candida. In the presence of chronic candidiasis, biopsy specimens of vaginal mucosa exhibit a less severe reaction.

2.6.1.2 Congenital infection

Candida infection acquired in utero and clinically manifested at birth is extremely rare. A review literature reveals that 10-35% of such cases have been reported(Darmstadt *et al.*,2000).

The obvious explanation is direct invasion of candida from the vagina to the amniotic fluid following premature or prolonged rupture of the membranes. Subclinical, self-healing ruptures in the membranes through which *C. albican* may enter the amniotic sac. Penetration of *C. albican* through intact membranes is a third possibility. This has been shown by the ability of *C. albicans* to infect and penetrate cluck chorioallantoic membrane and kill the embryo by invading the internal organs.

2.7 Diagnosis

Diagnosis depends on both demonstration of a species of candida, and the presence of clinical features compatible with the disease. Clinically, any patient with pruritus, redness or thrush-like patches on the vaginal must be suspected of having candidiasis. But, Donders *et al.*,(2002) have shown, none of these, single or in combination are good predictors of positive cultures. Unmistakable thrush patches on the vestibule or vagina should perhaps be considered pathognomonic. But at times, masses of epithelial cells from a physiological increase in desquamation are mistaken for thrush patches. Hyperkeratosis of the vaginal fornices, which occurs in some diaphragm users, may also be confused with thrush patches.

Using dipsticks, the pH of the vaginal secretion can be simply measured and it is a highly informative diagnostic test. When reliably obtained, a pH value equal to or less than 4.7 essentially limits the diagnosis to, either the patient has candida or she has a physiological discharge. In the presence of symptoms, the diagnosis is always candidiasis, which can then be confirmed by additional means.

2.7.1 Wet Mount Preparation

Microscope examination of relative thin vaginal material mixed with physiological saline, under low and high-power magnification, provides the most valuable single method for the differential diagnosis of vaginitis. The spores of *C. glabrata*are of variable size(2-8%μm) and spherical or ovoid in shape, mostly smaller than RBC. That of *C. albican* are uniform in size, isolated and almost always associated with hyphal filaments. The number of pus cells is variable. Predominance of lactobacilli, which is usually identifiable in the wet mount under high-power objective, in a case of vulvovaginitis with pruritus, is strong evidence of candidiasis (Ochei and Kolkatker,p.446).

2.7.2 Potassium Hydroxide (KOH) Preparation:

Microscope examination of vaginal smear mixed in 10%-20% KOH is the most efficient method for rapid identification of candida. In this solution, pus cells and red cells undergo rapid dissolution. The vaginal epithelial cells, skin scrapings with a high keratin content, rapidly clear from translucent to transparent, but a time, their outlines continue to appear as a type of ghost cell. The branching, budding hyphae of *C. albicans*, which are alkali resistant, stand out in sharp contrast, The spores of *C. glabrata* are also distinctively visible under low and high-power magnifications.

2.7.3 Stained smears:

Under the high power and oil-immersion lens of the microscope smear. Fixed and stained with methylene blue or other simple stains reveal vividly the structural features of candida. In gram stained smear, spores and conidia are strongly gram positive. The filaments are uniformly gram positive or have gram positive granules.

In the comparism made by Avwioro *et al* (2010) of smears and cultures for diagnostic efficiency, the Papanicolaou smear demonstrated mycotic in only 7.60%, culture was positive in 30.10%.

CHAPTER THREE

MATERIALS AND METHOD

3.1 Materials

Cytology records of all cervical smear diagnosed cases of candidiasis from the Histopathogy laboratory department in Federal Medical Centre. Laboratory number of patients was used for collection of the stained cervical smear. Cervical smear samples collected from women who came from routine screening

3.1.1 Sample collection

3.1.1.1 Materials require

An array spatula, preferably made of wood.

A glass slide frosted on one end, for patient's identification labelling

Plastic slide container [jar] containing fixative.

Sterile speculum.

3.1.1.2 Method of collection

The vaginal mucosa was carefully examined visualize for any lesion before using speculum to dilate it.

The speculum was then being introduced without any lubricant to expose the cervix

The wide end of the ayre spatula was used to scrape the ectocervix mucosa

Then the small end of the arye spatula was placed in the external os of the endocervical canal as deeply as possible and rooted at 360 gently scrapping the entire surface of the external os and the internal os.

The spatula was smear on a labelled slide

3.2 Fixation

The smeared slide was fixed immediately in 95% ethanol in a couplin jar for at least one hour (1hr).

3.3 Staining procedure

After fixation in 95% alcohol, smear was taken through descending grades of alcohol [80%, 70%, and 50%] for 1 minute each then to water.

Then stained in Harris haemotoxylin for about 5 minutes.

Rinsed in the tap water to stop the action of haematoxylin.

Rinsed in tap water.

The smear with rinsed in water.

The smears were differentiated in 1% acid alcohol until only the nuclei retain the stain (a few seconds).

The smears were rinsed in water and transferred to 70% alcohol for few seconds. Then to 95% alcohol for dehydration.

The smears were then stained in O.G .6 for 2 minutes.

The slide was rinsed in two changes of 95% alcohol.

Then stained in EA36 for about 2-4 minutes.

The slides were rinsed in two changes of 95% alcohol for a few second in each.

The smears were dehydrated in alcohol. Cleared in xylene and mounted in synthetic resin medium [eg DPX], and allowed to dry.

The smears were then examined microscopically, first with *10 0bjective to scan through the smears, for adequate smear content and then for candida and these were further confirmed with x40 objective.

CHAPTER FOUR

RESULT

4.1 Result interpretation

Out of the 400 cervical smear examined,only 8(2%) had *Candida species*. Out of the 8 samples positive for Candida, 8(100%) of them showed variability in their cell nuclear sizes,classic cell clumping and polymorphonuclear (PMN) Leucocyte background; 6(75%) showed a reactive intra-epithelial infiltration with PMN leucocyte;5(62.5%) have slight hyperkeratosis of some of their squamous epithelial cells.

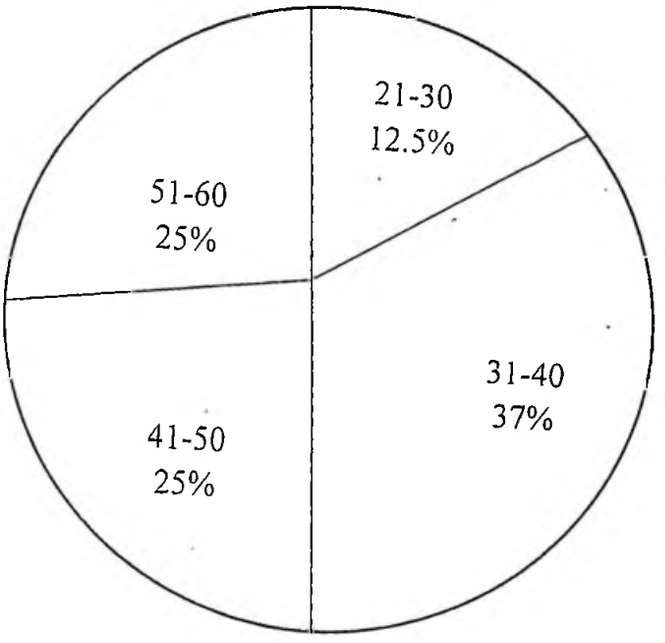
4.1.2 Table 1: Cellular changes associated with Candidiasis

S/No. cellular change	No. with the Change(%)	Prevalence(%)
Variability in cell Nuclear size	8	100
Reactive intra-epithelial Infiltration	6	75
With PMNL		
PMNL Background	8	100
Slight hyperkeratosis	5	62.5
Classical cell clumping Polymorphonuclear	8	100

$X^2 = 8.0627$

$P = 0.0893 (>0.05)$

It was also found from this study that 37.5% of those women within the age 31-40 had candida infection, while those who are 41-50 and 51-60 years had 25% rate of infection respectively.However,those within the age group 21-30 had the lowest rate of infection (12.5%) while in the groups 11-20 and 61 and above had no occurrence of infection.



Pie Chart showing prevalence of the infection in various age groups.

4.1.3 Cervical smear of candida infection morphology

Fig 4.1 A photomicrography of a smear with *Candida albican* infection x40. There are presence of pseudohyhae, inflammatory cells.

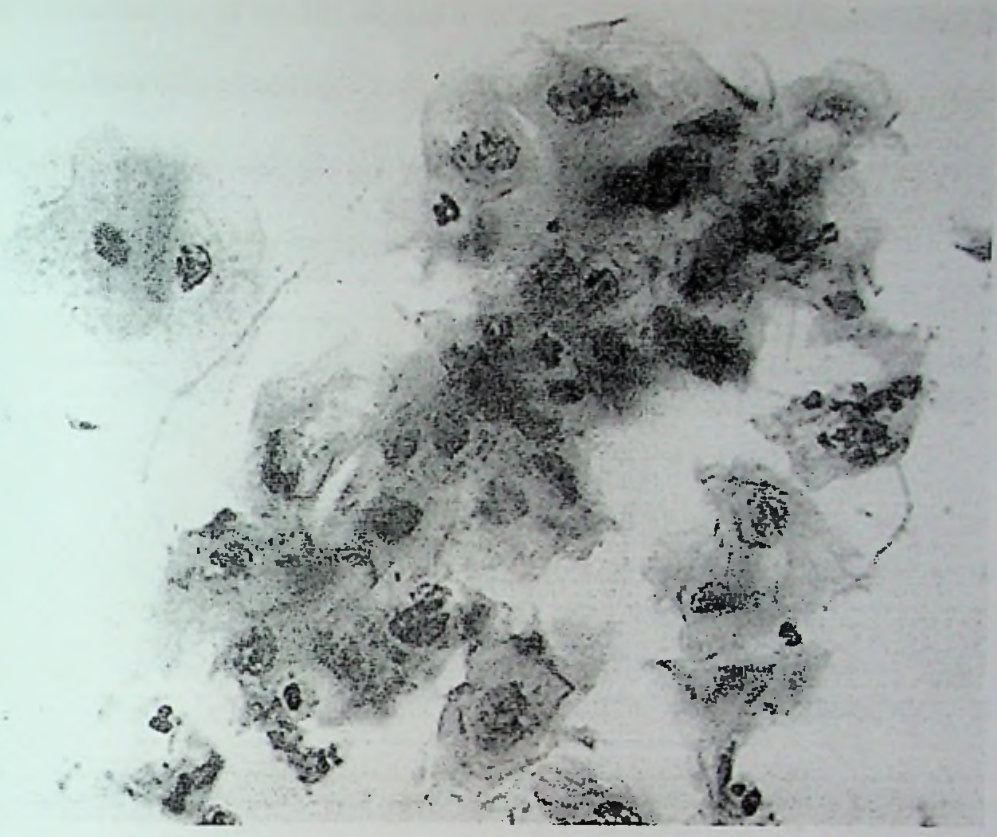


Fig 2:A photomicrograph of a smear of *Candida albicans* infection x40,

There are pink pseudohyphae, polymorphonuclear leucocytes at the background, superficial cells having vascular pyknotic nuclei and intermediate cells.

It was also found that out of the 382 women who were married, 7(1.75%) had candidiasis, while those who were single had only 0.25% prevalence of the infection. None of the widows had Candida infection.

4.1.4 TABLE 2: Effect of Marital Status

Marital status	No.	No. of positive	Prevalence (%)
Married	382	7	1.75
Single	11	1	0.25
Widow	7	0	0.00
Total	400	8	2

Six (1.5%) of the women with candidiasis had a history of recurrent vaginal discharge whilst 2(0.5%) of them had none.

4.1.5 Table 5: Recurrent vaginal discharge and candidiasis

Discharge	No.	No positive for	Prevalence (%) candida
Recurrent	263	6	1.5
Non recurrent	137	2	0.5
TOTAL	400	8	2.0

Out of the 200 women that used contraceptives devices, 8(1.25%) were positive for Candida while 3 (0.75%) of the remaining women with the infection used no contraceptive device (table 5).

4.1.6 Table 6: Effect of contraceptives

Use of contraceptives	No.	No. positive	Percentage(%)
Yes	200	5	1.25
No	200	3	0.25
TOTAL	400	8	2.00

Also, out of those who were married, those with a parity of 0-1 were discovered to have the highest rate of Candida infection (0.75%). This was followed by those with a parity of 4-5 (0.50%); While those with a parity of 2-3,6-7,10-11 had the lowest rate of infection.

Table 7: Parity and Candidiasis

Parity	No.	No. Positive for	Percentage(%)
	88	3	0.75
2-3	92	1	0.25
4-5	120	2	0.50
6-7	70	1	0.25
8-9	24	0	0.00
10-11	6	1	0.25
12 and above-			

CHAPTER FIVE

DISCUSSION RECOMMENDATION AND CONCLUSION

5.1 Discussion

Candidiasis is a depending pathogenic fungal infection caused by yeast genus Candida which comprises of about 154 species.20 of the species causes infection in humans. Candida colonies can grow rapidly within 3 days (*Candida albans et al*, 2015).

Candida specie (yeast) occasionally or habitually live on the skin and mucous membranes without causing harm to the host. Symptoms of candidiasis depend on the location of the mucosal lesion.

Higher prevalence of vaginal infection in all the age groups could be due to the fact that women are more sexually active and some of them have many sexual partners, whether they are married or not, in addition to the poor hygiene conditions. 'These results are in accordance with those of (Kamga *et al* 2014) . Candida specie (yeast) occasionally or habitually live on the skin and mucous membranes without causing harm to the host. Symptoms of candidiasis depend on the location of the mucosal lesion. The main study is to determine the prevalence of candidiasis and cytological changes among female patients presenting at Federal Medical Centre Keffi and to proffer possible solutions. It is assessed that 20% of ladies might be asymptomatically colonized by vaginal yeast (Sabel, 2007).

Out of the 400 cervical smear examined, 8(2%) had candida species. Out of the 8 samples positive for candidiasis, 8(100%) of them showed variability in their cell size, classic cell clumping and polymorphonuclear (PMN) Leuckocyte background; 6(75%) showed a reactive intra epithelial in filtration with PMN leucocyte, 5(62.5%) have slight hyperkeratosis of some of the squamous epithelial cell. 'the high prevalence of

these infections could be due to the anatomy of the female vaginal organ which favours penetration and the installation of the infection and poor personal hygiene. In fact, poor personal hygiene contributes to the change in pH from its normal state of acidity which could lead to the alteration of the normal flora; tight and synthetic underwear (in nylon) prevent aeration of the vagina, produce heat in the vaginal region, and then favour the proliferation of microorganisms. However, infected men naturally transmit the microorganisms to their partner due to the anatomy of female vaginal organ which acts like a reservoir. 'at is why 100% of women are generally infected The main study is to determine the prevalence of candidiasis and cytological changes among female patients presenting at Federal Medical Centre Keffi and to proffer possible solutions. It is assessed that 20% of ladies might be asymptomatically colonized by vaginal yeast (Sabel, 2007).

Candida albicans is commonly used as a model organism for pathogenic fungi (kabir *et al*, 2012). It is commonly referred to asdimorphic fungus as it grows both as yeast cells and filaments. However, it has several different morphological phynotypes (kadosh, 2019). Out of the 400 cervical smear examined, 8(2%) had candida species. Out of the 8 samples positive for candidiasis, 8 (100%) of them showed variability in their cell size, classic cell clumping and polymorphonuclear (PMN) Leuckocyte background; 6(75%) showed a reactive intra epithelial in filtration with PMN leucocyte, 5(62.5%) have slight hyperkeratosis of some of the squamous epithelial cell.

5.2 Recommendation

i. Since pregnancy, contraceptive use, abuse of antibiotics have been seen to contribute greatly in increasing the susceptibility to Candida infection, there is

the need for pregnant women to go for regular checkup in good hospitals with good laboratories.

ii. Furthermore, episodes should be promptly reported. For those who might need to carry out family planning, it is more advisable to sue the safe period method.

iii. It is also advice that all the women that are sent to the cytology laboratories for cervical screening should also have a swab collected for specific microbial investigation.

5.3 Conclusion

Vaginal organ infections are now gaining greater recognition as an important source of reproductive problems. Determining the aetiology and prevalence of these patients and their risk factors is important to establish the treatment strategy in the healthcare units. We therefore recommend that women should be routinely screened for vagina infections to reduce complications related to these infections.

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