Prevalence of cervical cytology abnormalities among pregnant women in Kenyatta National Hospital.

Dissertation submitted in part fulfillment of the requirements for Masters of Medicine in Obstetrics and Gynecology, University of Nairobi.

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DECLARATION

I declare that this research in part fulfillment of the M.Med degree in Obstetrics and Gynecology is my original work and has not been presented to any university forum.

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CERTIFICATE OF SUPERVISION

This is to certify that Dr. Samuel Wachira Ndungu researched upon this dissertation under my guidance and supervision and this book is submitted with my approval.

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DEDICATION

To my wife and friend, Liz Naini and our lovely daughter Tasha.

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To Almighty God, I give thanks and praise.

ABBREVIATIONS AND ACRONYMS

ANC	Antenatal clinic
ASC-H	Atypical squamous cell- cannot exclude HSIL
ASCCP	American society for colposcopy and cervical pathology
ASCUS	Atypical squamous cells of undetermined significance
CIN	Cervical intraepithelial neoplasia
CIS	Carcinoma insitu
DNA	Deoxyribonnucleic acid
ECC	Endocervical curettage
EGA	Estimated gestation age
FDA	United States food and drugs administration
HIV	Human immunodeficiency virus
HPV	Human Papilloma virus.
HSIL	High grade squamous intraepithelial lesion.
IARC	International agency for research on cancer.
IUCD	Intrauterine contraceptive device.
KNH	Kenyatta National Hospital.
LEEP	Loop electrosurgical excision procedure.
LSIL	Low grade squamous intraepithelial lesion
NARESA	Network for AIDS Research in East and South Africa.
NASCOP	National Aids and STD Control Programme.
Pap	Papanicolaou

PCR	**Polymerase chain reaction**
VIA	**Visual inspection with acetic acid**
VILI	**Visual inspection with Lugol's iodine**
VLPS	**Virus like particles**
WHO	**World health organization**

ABSTRACT

Background: Cancer of the cervix is the leading cause of cancer related deaths among women in Kenya. Cervical cytology screening programs have been shown to significantly reduce mortality and morbidity associated with cancer of the cervix. Since 90% of pregnant women in Kenya attend antenatal clinic (ANC) at least once, the ANC offers an excellent opportunity for cervical cytology screening.

Objective: To determine the prevalence of abnormal cervical cytology among pregnant women attending ANC at Kenyatta National Hospital (KNH).

Design: Cross-sectional descriptive study.

Setting: Kenyatta National Hospital antenatal clinic.

Study population: Pregnant women attending the first (booking) antenatal visit from February 2008 to August 2008.

Methods: A Papanicolaou (pap) smear was taken for each consenting participant during the first antenatal visit. A cervex™ brush was used to collect the cervical smears. The specimens were analyzed and reported on a standardized laboratory form. The social demographic profiles of the participants were obtained using a structured questionnaire. HIV serostatus was obtained from the participants' routine antenatal profiles.

Results: A total of 171 women attending their first ANC visit were recruited. The mean maternal age was 28.5 years with a range of 18 to 42. The mean gestation at the booking visit was 28 weeks with 57.9% of the participants initiating ANC in the 3rd trimester. Only 10.5% had had previous screening for abnormal cervical cytology. One hundred and sixty six (97.1%) pap smear specimens were satisfactory for evaluation. Five (2.9%) were unsatisfactory for evaluation. The prevalence of Squamous intra-epithelial lesions was 5.8% (3.5% LSIL, 2.3% HSIL) . Thirty one percent were inflammatory, 1.2% had Trichomonas vaginalis, 7% had bacterial vaginosis (diagnosed by the presence of clue cells) and 10.5% had Candida spp. Forty nine percent of the pap smears were reported normal. HIV sero-positive status and abnormal vaginal discharge were significantly associated with the presence of any pap smear abnormality (p value <0.05).

Conclusion

The prevalence of abnormal cervical cytology was 5.8%.

Positive HIV sero-status and presence of abnormal vaginal discharge were noted to be associated with a higher risk of pap smear abnormalities.

The rate of previous screening for cervical cytology was low. Only 10.5% of the participants had had a pap smear done in the past.

Recommendation

Cervical cytology screening should be included in routine antenatal care in our hospitals. This would be in line with the WHO model on focused antenatal care which recommends cervical cytology screening on the first antenatal visit.

Antenatal care givers should actively promote and provide counseling and testing for HIV as this was observed to increase the risk of abnormal cervical cytology.

Antenatal mothers with vaginal discharge should be evaluated to rule out reproductive tract infections.

1.0 Introduction

Cancer of the cervix is the most common reproductive tract malignancy in Kenya. Data from the Nairobi Cancer Registry, a population based cancer registry at the Kenya Medical Research Institute, indicate that cervical cancer accounted for 18 to 23% of all malignancies reported between the year 2000 and 2003.

Despite the morbidity and mortality associated with cervical cancer, it is estimated that only 1% of Kenyan women have been screened for cancer of the cervix, compared to 70% in the developed world (1).

The pap smear test is one of the most successful strategies developed for prevention of cancer. It has been credited with more than 70% reduction in cervical cancer related deaths in the United states between 1948 and 1988 (2, 3).

According to the Kenya Demographic and Health survey 2003, 90% of pregnant women attended Antenatal clinic at least once. Therefore the ANC offers an excellent opportunity for cervical cytology screening.

The local prevalence of abnormal cervical cytology in pregnancy has not been documented. In the United States, 2% to 7% of pregnant women have an abnormal pap. Cervical neoplasia (including carcinoma insitu and invasive carcinoma) is estimated to complicate 1.5 to 12 of every 100,000 pregnancies (4, 5).

The evaluation of abnormal cervical cytology during pregnancy can be complicated by cervical physiological changes such as increased vascularity and hyperplasia of endo-cervical glands. The choice of treatment modalities can also present a

dilemma for the gynecologist. The goal of screening during pregnancy should be to exclude invasive disease.

1.1 Literature Review

Cancer of cervix is the 2nd leading cause of cancer deaths in women worldwide with more than 270000 deaths reported every year. Over 80% of these deaths occur in developing countries, where cervical cancer is the leading cause of cancer death (6).

The International Agency for Research on Cancer has estimated that the age-standardized incidence of cervical cancer in Kenya was 36.6 per 100,000 women in the year 2000, which translates to nearly 3,000 new cases per year(1).

The local prevalence rates of cervical dysplasia in non pregnant women range from 2.56% to 14%. In rural Kenyan populations, Ndavi, Kitavi and Chamia in separate studies found very consistent prevalences of 2.56%, 2.6%, and 2.64% respectively (7, 8, 9). The three investigators used the CIN reporting system. A study of women attending a family planning clinic in Nairobi Kenya found a prevalence of 12% for cervical dysplasia (mild in 5.8%, moderate in 3.5% and severe in 1.2%). 1.5% had invasive cervical cancer(10). A similar study of 429 women attending a family planning centre in Nairobi between 1998 and 2000 found a 14% prevalence of dysplasia,(7% had LSIL, 6.8% had HSIL) while 0.23% had invasive cancer (11).

There are no local studies done to determine the prevalence of abnormal cervical cytology in pregnant women.

The risk factors for cervical dysplasia are similar to those for cancer of the cervix. They are linked to sexual exposure and include early onset of coitus, multiple sexual partners, multiparity and exposure to sexually transmitted infections especially Human Papilloma Virus. Recent studies have demonstrated that with the probable exception of a very rare type of highly differentiated squamous cell carcinoma, Human Papilloma Virus (HPV) is responsible for 99.7% of all invasive carcinomas of the cervix (12, 13). Genital HPV types have been classified into categories of low oncogenic risk (e.g. types 6 and 11) and high oncogenic risk (e.g. types 16 and 18). Infection with high oncogenic risk types is necessary for the development of precancerous neoplastic lesions and/or cancer of the cervix. A survey of HPV types in invasive cervical cancer in 22 countries around the world found that HPV 16 accounted for 54% of cancer of the cervix followed by HPV 18(15%), HPV 45(9%), and HPV 31(6%). Thus these four HPV types accounted for 84% of all cases of cancer of the cervix (14, 15).

HIV infection is also a strong risk factor for cervical cancer, with HIV sero-positive women 5 times more likely to have squamous intraepithelial lesions and lower genital tract neoplasia than HIV sero-negative women (16). Other risk factors include family history of cancer of the cervix, long-term use of oral contraceptives and cigarette smoking.

Human Papilloma Virus vaccines have been studied for primary prevention of cancer of the cervix. Two vaccines against the most common cancer causing HPV types have been developed and tested in clinical trials and are now licensed (17-22). Both vaccines are based on the recombinant expression and self assembly of the major capsid protein L1 into virus like particles (VLPS) that resemble the outer capsid of the HPV virus. They contain no viral DNA and are not live/attenuated viruses. Injection of the HPV VLPS elicits a strong and sustained type specific response (23, 24). One of the vaccines, Gardasil (Merc & co. Inc) protects against HPV types 6, 11, 16 and 18(quadrivalent) while Cervarix (GlaxoSmithKline) protects against types 16 and18 (bivalent). Only Gardasil has FDA approval and hence forms the basis for American Cancer Society vaccine guidelines;

- Routine vaccination is recommended for girls 11 to 12 years.
- Girls as young as 9 years may receive HPV vaccination.
- HPV vaccination is also recommended for females aged 13 to 18 years to catch up missed vaccine or complete vaccination series.
- There is currently insufficient data to recommend for or against universal vaccination of females aged 19 to 26 years in the general population. A decision can be made after an informed discussion between the woman and her health provider. Ideally the vaccine should be administered prior to potential exposure to genital HPV. The potential benefit is likely to diminish with increasing number of lifetime sexual partners.

- HPV vaccination is not currently recommended for females above 26 years or for males.
- Screening for cervical intraepithelial neoplasia should continue for both vaccinated and non-vaccinated women.

Several randomized controlled trials have shown that both the bivalent and quadrivalent vaccines prevent persistent HPV and HPV related CIN 2/3 (18, 19, 21, 22). The two vaccines showed an efficacy of close to 90%. However the enrollment criteria for the vaccine trials restricted the number of lifetime sexual partners to a maximum of four. When subjects entered the studies with evidence of current or past HPV infection (i.e. PCR or serology positive), there was no clear evidence of protection from subsequent disease (25).

There is currently no information about the duration of vaccine induced immunity. Hence long-term post licensure studies are needed.

The Pap smear has been used for over 50 years as a screening test for pre malignant lesions of the cervix. Its effectiveness has been demonstrated in the developed countries where with cervical cytology screening programs, there has been a direct relationship between the proportion of population screened and declining incidence of cervical cancer. However, the Pap smear does not establish the diagnosis of cervical cancer .The false negative rate of the Pap test for cervical cytology screening is thought to be 15% to 25%; however a recent review by the Agency for Health Care Policy and Research found a much lower sensitivity of the convectional Pap smear at 50%(26).

Liquid based cervical cytology preparations have been shown to improve on the sensitivity of the Pap test. They improve on cell transfer from the collecting instrument. The thin layer slide reduces obscurity and produces uniformity of the cell sample (27, 28). Several organizations including The American Cancer Society recommend HPV DNA testing in conjunction with cytology as a screening option for females above 30 years. Colposcopy and cervical biopsy are however still required for diagnosis.

Pap smear specimens can be obtained using the cytobrush and spatula technique or the cervex™ brush (broom). The cervix is first exposed using a Cusco's speculum. A sample is obtained from the endocervix using the cytobrush by rotating through 90 to180 degrees. A wooden spatula is then used to obtain samples from the transformation zone and the ectocervix (26, 29).

In the cervex brush technique, the central bristles of the brush are inserted in the endocervical canal while the lateral bristles are bent against the ectocervix. The cervex brush is then rotated five times while maintaining gentle pressure. The sample is then transferred to the slide via painting like movements on each side of the brush. A fixative is then applied on the slide.

A cotton brush and spatula have previously been used to obtain pap smears but was found to be unlikely to avail endocervical cells for cytological evaluation. Use of cytobrush and cervex brush in pregnancy has been shown to be more likely to obtain endocervical cells with no increase in both minor and major complications (30, 31, 32).

According to the American College of Gynecologists, cervical cytology screening should be started three years after initiation of sexual activity or at the age of 21 years. Annual cytology screening is recommended for women below the age of 30 years. Women aged 30 years and above who have had three consecutive cervical cytology reports that are negative for intraepithelial lesions or malignancy may be screened every 2 to 3 years. However, women with other risk factors e.g. immunosuppression require more frequent screening (33). Women aged 70 years or more may elect to stop cervical cytology screening if they have had three consecutive normal test results and no abnormal Pap smears results within the prior 10 years.

In Kenya, The ministry of health, through the division of Reproductive health, is in the process of implementing The National Cervical Cancer Prevention Programme (Action plan 2005-2009) (1). The objectives of the programme are:

- ✓ Make screening and pre-cancer treating services available in at least 20 districts by the year 2009.
- ✓ Mobilize high risk women (30-49 years old and never been screened before) to go for pre-cancer screening. Sustain the cancer prevention programme so that repeat screening can be done after five years.
- ✓ Ensure that at least 70% screen positive women receive appropriate care within 3 months of being screened.

- ✓ Strengthen at least three regional referral facilities (western, central and eastern areas) for management of cervical cancer patients, including diagnosis, surgery and palliative care.

The service model for the National Cervical Cancer Programme is to be integrated into existing reproductive health services (including maternal child heath and family planning departments). This would make them more acceptable and cost effective than parallel screening programmes.

Visual inspection with acetic acid (VIA) and Visual inspection with Lugol's iodine (VILI) have been proposed as the primary cervical cancer screening methods for the national programme. However private physicians may continue to offer pap smears for cervical cytology screening.

Interpretation of cervical cytology in pregnancy can be complicated by the physiological changes of pregnancy. Michael and Esfahani conducted a retrospective review of 278 abnormal Pap smears in both pregnant and post partum women. Several features were identified that may make the differentiation of normal versus abnormal cervical smears in pregnancy difficult. They reported an abundance of degenerated decidual cell which may mimic high grade squamous epithelial cells (HSIL). Cytotrophoblasts distinguished only by the presence of prominent nucleoli may also mimic HSIL. Syncytiotrophoblasts may be confused with Human Papilloma Virus (HPV) effect based on perinuclaer cavitations and nuclear atypia. Cells exhibiting Arias-Stella reaction with

vacuolated cytoplasm and enlarged atypical nuclei may imitate cytological abnormalities associated with endocervical adenocarcinoma (34).

The question of adequacy is also relevant. Londo et al conducted a retrospective review of 1377 obstetric patients. Endocervical cells were present in only 41% of the prenatal samples compared to 82% of the post partum smears. However the findings could be explained by the method used for Pap smear collection. A cotton swab Ayre spatula method was used in pregnant women while the cyto-brush was used in non pregnant women (35).

With the 2001 Bethesda system, smears not containing an endocervical component are no longer termed "inadequate" or "limited". However the lack of endocervical cells is still mentioned in the cytology report.

Although cytological interpretation is subject to the physiological effects of pregnancy, any suscipicious appearing cells should be evaluated.

Colposcopy is the principle diagnostic procedure for evaluation of abnormal cervical cytology in pregnancy. Colposcopy in pregnancy is safe, reliable and can be done regardless of gestation age (36).

The colposcopic appearance of the gravid cervix differs markedly from the non gravid cervix. The physiological changes include cervical hypertrophy, ectopy/distal migration of the Transformation zone and redundancy of the hyperemic vaginal mucosa. The Squamo-columnar junction everts and is completely visible by 20 weeks gestation (36). Hence if colposcopy is unsatisfactory in early pregnancy, it can be repeated later in pregnancy.

Increased cervical perfusion may cause aceto white epithelial changes to appear less prominent. This change may mask a High Grade Squamous Lesion and make it appear like a Low Grade Squamous Lesion. Additionally, as physiological eversion continues in pregnancy, areas of squamous metaplasia may become more apparent, and can also appear aceto white on application of acetic acid during colposcopy.

Increased laxity of the vaginal wall may obscure colposcopic view of the cervix. Gentle retraction with a wooden tongue spatula may facilitate visualization.

Diagnosis of early invasive cancer may be delayed due to a physician's reluctance to perform colposcopically directed punch biopsies of the cervix in pregnant women. Performance of punch biopsies is safe in all trimesters and is not associated with any increased risk of bleeding compared to non pregnant patients, although many colposcopists will defer punch biopsy until second trimester. Bleeding is generally controlled via direct pressure or application of nitric acid (37, 38, 39).

Endocervical curettage (ECC) endangers the pregnancy and is therefore contraindicated in pregnancy (37, 38, 40). However, the use of cyto-brush to sample the endocervix has been reported to be safe in pregnancy (31, 41).

Management of Abnormal Cervical cytology in pregnancy depends on the cytological or histological findings. The guidelines are provided by the American Society for Colposcopy and Cervical Pathology (ASCCP) (40).

For pregnant women with atypical squamous cells of undetermined significance, ASCCP recommends that these women be managed similarly to non pregnant women, i.e. the Pap smear is repeated in six months or after six weeks post partum (40).

Colposcopy is indicated for women with Atypical Squamous cells – Cannot rule out a High Grade Lesion (ASC-H), LSIL and HSIL, Atypical glandular cells and Adenocarcinoma insitu.

Unless invasive disease is suspected, pregnant women may be followed up with cytology and colposcopy for the remainder of pregnancy, with biopsy if the lesion appears to worsen or if cytology report is suggestive of invasive cancer (40).

In the event that pre-invasive disease is confirmed by colposcopy and or cervical biopsy, conservative management and surveillance during pregnancy is the expected standard of care (5, 39, 42, 43). For the pregnant patient with CIN, it is reasonable to evaluate the patient with cytology every 8 to 12 weeks. If indicated definitive treatment could be completed at 6 to 8 weeks post partum.

The effect of pregnancy on the natural history of cervical dysplasia is controversial. Although several studies demonstrate no association between pregnancy and post partum regression rates of moderate to severe dysplasia (43, 44), others describe a high incidence of regression by the time of post partum evaluation (39, 45, 46, 47). In one study of 138 women with abnormal ante partum cervical cytology, Adhoot et al reported that the over all regression rate of

ASCUS was 65%, the regression rate for LSIL was 64% and 47% for HSIL (48) The study did not comment on rates of progression to a higher grade of dysplasia. An Australian study which included 811 women with abnormal cervical cytology found similar regression rates to Adhoot et al, but also noted a 7% progression rate to a higher grade of dysplasia. No progression to invasive carcinoma was reported (49).

The evidence therefore supports observation for most patients with abnormal cervical cytology in pregnancy. However if results of either the colposcopic or histologic examination are suggestive of invasive disease, a diagnostic excision procedure is indicated.

Cervical conization in pregnancy has been associated with significant blood loss (~500mls) in approximately 10% of patients. Other complications include preterm labor and abortion rates of up to 18% in first trimester (37). Conization may be modified to obtain a shallower specimen, i.e. a "coin biopsy" to decrease the likelihood of adverse effects. This may however yield a specimen with positive margins and place the patient at high risk of residual disease (50, 51).

Several authors have suggested that if conization is considered in pregnancy, cervical cerclage should be performed concurrently (52). A MacDonald cerclage is placed prior to conization and tied immediately at the completion of the procedure.

Loop electrosurgical excision procedures have been studied in pregnancy. There is no evidence of an advantage of LEEP compared to Cold-knife cone biopsy

the complications of prematurity (56,57). It is generally recommended that these women deliver via caesarian section although even with invasive cancer the route of delivery appears to have no impact on the fetal or maternal outcome (57). Radical hysterectomy and pelvic lympadenectomy can be performed concurrently with the caesarian section. For patients with EGA between 20 and 24 weeks, management decisions should be made after the mother has been counseled on the involved maternal and fetal risks. Counseling should be done in conjunction with a neonatologist and a gynecologic oncologist.

with respect to positive margins or hemorrhage when performed on pregnant women (53, 54). Robinson et al demonstrated that LEEP performed during pregnancy does not consistently produce diagnostic specimens and is associated with a significant rate of residual disease (47%) identified during post partum evaluation (54). More over the associated morbidity appears to be similar to that associated with cone biopsy and is more common in the 3rd trimester. Robinson et al concluded that LEEP during pregnancy should be reserved for the same indications as cone biopsy (54).

Management of invasive cervical carcinoma in pregnancy depends on the gestational age, stage of disease, and maternal desire to continue with the pregnancy. Most patients diagnosed with cervical cancer in pregnancy have stage one disease. Approximately 76% are diagnosed as stage IB and 78% have squamous cell carcinoma (55). If stage IA1 (<3mm of stromal invasion) disease is diagnosed on conization with clear margins, it is reasonable to follow the pregnancy until term and anticipate a vaginal delivery. For patients with advanced disease with EGA less than 20 weeks, the recommendation is to treat without delay. Treatment may include radical hysterectomy with bilateral pelvic lympadenectomy or radiation therapy depending on the stage of the disease (56). If radiation therapy is to be used, external beam radiation can be initiated. The pregnancy will typically abort during therapy (56, 57). For those with an EGA greater than 24 weeks its reasonable to wait until term or near term taking into account the availability of neonatal support to optimize fetal well being and avoid

the complications of prematurity (56,57). It is generally recommended that these women deliver via caesarian section although even with invasive cancer the route of delivery appears to have no impact on the fetal or maternal outcome (57). Radical hysterectomy and pelvic lympadenectomy can be performed concurrently with the caesarian section. For patients with EGA between 20 and 24 weeks, management decisions should be made after the mother has been counseled on the involved maternal and fetal risks. Counseling should be done in conjunction with a neonatologist and a gynecologic oncologist.

2.0 Rationale

The new World Health Organization model for antenatal care recommends cervical cytology screening during the first antenatal visit. In Kenya it is not routine to screen for cervical cancer during pregnancy and therefore there are no studies done to determine the prevalence of abnormal cervical cytology in pregnancy.

Since 90% of pregnant women in Kenya attend antenatal clinic at least once, the antenatal period offers an excellent opportunity for cervical cytology screening. This would go along way in reducing morbidity and mortality associated with cancer of the cervix.

Cancer of the cervix is the most common gynecological cancer in Kenya. It accounts for most of the cancer related mortality in developing countries. World wide, both the incidence and mortality of cancer of the cervix are only second to cancer of the breast. Elaborate cervical cytology screening programmes have been shown to markedly reduce morbidity and mortality associated with cancer of the cervix.

Study questions

1. What is the extent of abnormal cervical cytology among pregnant women attending ANC at Kenyatta National Hospital?
2. What are the factors associated with abnormal cervical smears?

2.1 OBJECTIVES

2.1.1 Broad Objective.

To determine the prevalence of abnormal Pap smears among pregnant women attending Antenatal clinic in Kenyatta National Hospital.

2.1.2 Specific Objectives

1) Describe cervical cytology findings among women attending ANC in KNH.
2) Determine the specimen adequacy of pap smears collected during pregnancy.
3) Determine correlates of abnormal cervical cytology.

3.0 METHODOLOGY

3.1 Study Design

This was a hospital based cross-sectional descriptive study, carried out over a period of five months. The cross-sectional design is the most appropriate in a prevalence study.

3.2 Setting

The study was done at the Kenyatta National Hospital's ANC which serves the population within and around the city. A number of the antenatal clients are initially seen at peri urban peripheral hospitals and are referred to KNH. KNH is a national referral and a teaching hospital for the University of Nairobi and the Kenya Medical Training College. The obstetrics unit consists of three

antenatal/postnatal wards, labor ward, a maternity operating theatre, antenatal and post natal clinics.

About 50 to 60 new mothers are seen on every Monday booking clinic.

The obstetrics department is currently implementing the new WHO focused antenatal care model. Four antenatal visits are recommended for antenatal mothers with no pregnancy associated complications. Mothers with high risk pregnancies are however followed more closely. The WHO model recommends only one routine vaginal examination during pregnancy, which includes taking a pap smear if the patient has not done one during the past two years.

3.3 Study population/study period

The study population included pregnant women attending their first antenatal visit at Kenyatta National hospital, from February 2008 to August 2008. The investigator opted for mothers attending their first antenatal clinic in order to comply with the WHO guidelines on focused antenatal care which recommend cervical cytology screening on the first antenatal visit. Participants were recruited on the Monday booking clinic as well as new patients on Tuesday, Wednesday and Thursday antenatal clinics.

3.4 Sample size

The sample size was calculated using the following formula (Woolson 1987).

$$n=\frac{Z^2PQ}{d^2}$$

Where n = Desired sample size

Z = Reliability coefficient at δ (0.05) level of significance

=1.96 at δ of 0.05.

P = Prevalence; a prevalence level of 12% was assumed (A study done in a family planning clinic in Nairobi found a prevalence of 12% [10].

Q = 1.0 – p

d = Degree of precision (5%)

$$n = \frac{(1.96)^2 x0.12 \times 0.9}{(0.05)^2}$$

$$n = \underline{165}$$

A total of 171 participants were recruited for the study.

3.5 Inclusion Criteria

- Pregnant women attending their first antenatal visit at Kenyatta National Hospital.

3.6 Exclusion Criteria

- Women with the following complications:
  - Early pregnancy bleeding.
  - Ante partum hemorrhage
  - Preterm premature rupture of membranes, preterm labor or obvious cervical dilatation on speculum exam.
- Observed florid vaginal discharge.

3.7 Study instruments

A coded questionnaire and a standardized laboratory request form were used.

3.8 Study procedure

Recruitment was done on all the antenatal clinic days (Monday to Thursday). Consecutive sampling was done where every antenatal mother who met the inclusion criteria was recruited, until the desired sample size was achieved. The investigator with the assistance of a trained study nurse first reviewed the files of the antenatal clients as they came to the observations area. Those who satisfied the study criteria were informed of the study and those willing to participate were considered eligible. Eligible clients were called one by one into the consulting room. The investigator went through the information on the consent form regarding the relevance of the study, risks and discomfort, questionnaire, physical examination and specimen collection. Willing clients then signed the consent form.

A coded questionnaire was administered to extract socio-demographic data and past obstetric and gynecological characteristics.

The participants were provided with additional counseling to ensure they were comfortable prior to speculum examination and pap smear collection. The investigator then cleaned hands, adorned a pair of sterile latex gloves and in the presence of a nurse chaperon the mother was requested to lie in semi lithotomy position. The investigator then parted the labia with the left hand and gently introduced the speculum. The gross appearance of the cervix and any abnormal vaginal discharge was noted and recorded in the laboratory request form. A Pap smear was then obtained for each participant. A **Cervex**™ brush was used to obtain the cervical smear specimen by gently rotating it four to five times against the ectocervix. The sample was immediately transferred to a slide and fixed with 95% ethyl alcohol. The participants were given a four week return date for review with results. All the fixed specimens were then taken on the same day to University of Nairobi pathology department for processing and reporting. The staining was initially done with haematoxyllin, differentiated with 0.05% acid water and dipped in blue Scott's solution for one minute. Rinsing was done with 95% ethyl alcohol. The staining was completed with orange green 6 and Eosin azure stains and then mounted for screening. The Bethesda system (2001) was used for Pap smear screening with a standardized form used for reporting each specimen. A copy of the result was filed in each participant's antenatal file. The reports were availed to the participants by the investigator. Participants with

infections e.g. bacterial vaginosis, candidiasis and trichomoniasis were given relevant prescriptions. Those with abnormal cervical cytology i.e. LSIL and HSIL were immediately referred to the colposcopy clinic within the same building for management by gyne-oncologists.

3.9 Quality control/result validation

Initial cytological evaluation for all the smears was done by a cytopathologist from University of Nairobi. For quality control and results validation, a second evaluation by an external cytopathologist was done for randomly selected pap smears and for all the smears with initial abnormal results. There was a hundred percent concurrence between the two cytopathologists.

3.10 Data collection and management

Data was collected using a coded questionnaire and a standardized laboratory request form. Questionnaires and laboratory report forms were kept under lock and key prior to and after data entry. During entry, data was backed up daily on a DVD and kept by the investigator. Once data entry was complete, data cleaning was done by checking each entry against the hard copy forms. Data was then exported from the data base and analyzed using Statistical Package for Social Sciences (SPSS 12) for Windows and S-Plus. Data analysis involved descriptive statistics, like cross tabulation, frequency, ranges, mean. The prevalence of cervical cytology abnormalities was estimated by calculating the proportion of study participants with abnormal cytology results i.e.

Prevalence of abnormal cervical cytology = $\frac{\text{no. of clients with abnormal cytology}}{\text{Total Sample size}} \times 100$

$$= \frac{10}{171} \times 100 = 5.8\%$$

Data on quality of pap smears was extracted from the standardized laboratory request form. Correlation between socio-demographic and obstetric/gynecological characteristics with cervical cytology abnormalities was evaluated using Pearson's Chi-square test. Fisher exact test was used when appropriate. Significance was defined as P value <0.05. Multivariate analysis was done for all the statistically significant variables.

3.11 Study limitations

1) The data collected was hospital based and KNH being a referral centre results may not be generalizable to the general population.

2) Participant's recent history of douching, recent sexual activity and antibiotic use may not have been revealed and documented.

3) Participants with inflammatory pap smear results did not have repeat pap smears after treatment. This was largely due time limitation.

4) Recall bias could have arisen where clients were asked about their past obstetric and medical history. Attempts were made to make the questionnaire clear and translation done where necessary.

5) The physiological changes that occur on the cervix during pregnancy could have compounded cervical cytological reporting. A second evaluation for randomly selected smears and for all smears with abnormal cytology was however done by the quality control cytologist.

3.12 Ethical considerations

The study was approved by the ethical and research committee of Kenyatta National Hospital. Voluntary informed consent was obtained from every participant prior to administration of the questionnaire and Pap smear collection.

Clients who declined to participate in the study received the standard KNH antenatal care. Their services were not affected in any way.

Results of Pap smear cytology were availed to all participants. Participants with abnormal results were treated appropriately. Those requiring colposcopy were referred to KNH colposcopy clinic.

Data and information was treated with confidentiality. The participant's names and ANC number did not appear on the questionnaire or the laboratory request form. A serial number was used instead.

Results of the study will be made available to KNH department of Obstetrics and Gynecology.

4.0 RESULTS

4.1 Socio-demographic characteristics.

A total of 171 women attending their first (booking) antenatal visit were recruited. The mean maternal age was 28.5 years (SD±5) with a range of 18 to 42 years.

Table 1, Socio-demographic characteristics of the participants.

Socio demographic characteristics	Frequency (%) N=171
Education Level	
Primary	28 (16.4%)
Secondary	53 (31%)
University/college	90 (52.6%)
Marital status	
Single	21 (12.3%)
Married	150 (87.7%)
Occupation	
Unemployed	61 (35.7%)
Employed (casual)	8 (4.7%)
Self employed	42 (24.6%)
Long term employment	60 (35.1%)

More than 87% of the participants were married. Majority, (52.6%) were either in college or had attained college/university level of education. Thirty five percent were unemployed while almost an equal number were in skilled long term employment.

4.2 Obstetric and gynecological characteristics

The mean gestation age on the first antenatal visit was 28 weeks (SD ±8.7) with a range of 5 to 40 weeks. Ninety nine (57.9%) of the participants came for their first antenatal visit in the third trimester.

Table 2, Obstetrics and Gynecological characteristics of the participants.

Obstetric & gyne characteristic	Frequency (%) N=171
Mothers' parity	
Primigravida	74 (43.2%)
Para 1	60 (35.1%)
Para 2	26 (15.2%)
Para 3 and above	11 (6.4%)
Age at Sexual Debut	
10-14	3 (1.8%)
15-19	83 (48.5%)
20-24	70 (40.9%)
25-29	13 (7.6%)
30+	2 (1.2%)
Lifetime Sexual partners	
1	52 (30.4%)
2	63 (36.8%)
3	34 (19.9%)
4	15 (8.8%)
5 and above	7 (4.1%)
Method of contraception prior to current pregnancy	
None	80 (46.8%)
Natural Methods	4 (2.3%)
Barrier Methods	5 (2.9%)
Injectable Progestin	25 (14.6%)
Progestin Implants	9 (5.3%)
Combined oral pill	43 (25.1%)
Intrauterine Devices	2 (1.2%)
History of STIs'	
Yes	16 (9.4%)
No	155 (90.6%)
Pap Smear done Previously	
Yes	18 (10.5%)
No	153 (89.5%)

Most the participants were of low parity. Forty three percent were primigravidas while 35% were para 1. The age at sexual debut was low with 50% having had sexual contact before 19 years of age. Thirty percent had one lifetime sexual partner, while close to 37% had two lifetime sexual partners. Only 4.1% had more than five lifetime sexual partners. The over all contraceptive uptake prior to the index pregnancy was 53%. Twenty five percent had used combined oral contraceptive pill. IUCD was least popular at 1.2%.

Out of 171 participants, only 18(10.5%) reported having done a pap smear previously.

4.3 Other risk factors for abnormal cervical cytology

Table 3, other risk factors associated with abnormal cervical cytology.

Other Risk Factors for Abnormal cervical cytology	Frequency (%) N=171
Cigarette Smoking	
Yes	6 (3.5%)
No	155(90.%)
HIV Status	
Negative	157(91.8%)
Positive	14(8.2%)

Fourteen (8.2%) of the participants were HIV sero-positive while 3.5% smoked cigarettes.

4.4 Pap smear Quality

A total of 171 pap smears were evaluated. One hundred and sixty six (97.1%) of the pap smears were satisfactory for evaluation.

Table 4, Quality of cervical smears.

Specimen satisfactory for evaluation but partially limited by;	**Frequency (%)**
Excess blood cells	6 (3.5%)
Lack of endocervical cells	29(17.0%)
Excess inflammation/pus cells	42(24.6%)

Six (3.5%) of the pap smears were partially limited by excess blood cells. Seventeen percent were noted to lack endocervical cells while 24.6% were noted to have excess pus/inflammatory cells. **Five (2.9%)** of the specimens were inadequate for evaluation.

4.5 Pap smear results

Table 5, pap smear results		N=171
Abnormal		**96 (56.1%)**
Squamous cell abnormalities	**10 (5.8%)**	
LSIL	6 (3.5%)	
HSIL	4 (2.3%)	
Infections/inflammation	**86 (50.3%)**	
Inflammatory smears	54 (31.6%)	
Trichomonas vaginalis	2 (1.2%)	
Bacterial Vaginosis	12 (7.0%)	
Organism consistent with Candida	18 (10.5%)	
Normal		**70 (40.9%)**
Inadequate for evaluation		**5(2.9%)**

Ninety six (56%) of the pap smears were abnormal. Of the abnormal smears, ten (5.8%) had squamous intraepithelial lesions. There was a high prevalence of inflammatory smears (31.6%) with no specific organism identified. The prevalence of Trichomonas vaginalis was low at 1.2% while bacterial vaginosis was more common at 7.0%. The most common infection identified was candidiasis at 10.5%.

Seventy (40.9%) of the pap smears were normal. There were no glandular cell abnormalities. Five pap smear specimens (2.9%) were inadequate for evaluation.

4.6 Association between socio-demographic, obstetrics/gynecological and clinical characteristics with any cervical smear abnormality.

Out of 171 pap smears evaluated, there were only 10 intraepithelial lesions. It would not be statistically appropriate to compare this small number with all the other smears that were negative for intraepithelial lesions. Hence, correlation was done with any abnormal cervical smears (including benign cellular changes and infections).

Table 6, Correlation between Age and any Pap smear abnormality.

	Over all N=171	Any pap smear abnormality N=96	Normal pap smear N=70	P value
Mean maternal age	28.5 (18-42)	27.7(18-40)	29.3(20-42)	0.068

The mean age for mothers with abnormal pap smears was 27.7 years while that of mothers with normal pap smears was 29.3 years. There was no statistically significance association between age and the presence of any pap smear abnormality (p=0.068).

Table 7, correlation between any Pap smear abnormality and obstetric/gynecological characteristics.

Variable	Overall N=171	Any pap smear abnormality	Normal pap smear	P value
Number of births (parity)				
None		44(25.7%)	30(17.7%)	
1		35(20.5%)	25(14.6%)	0.65
2		16(9.4%)	10(5.8%)	0.95
3 and above		6(3.5%)	5(2.9%)	0.50
Age of sexual debut				
10-14		1(0.6%)	2(1.6%)	0.34
15-19		50(29.2%)	33(19.3%)	0.87
20-24		43(25.1%)	27(15.8%)	
25-29		6(3.5%)	7(4.1%)	0.30
30+		1(0.6%)	1(0.6%)	0.74
Lifetime sexual partners				
1		34(20%)	18(10.5%)	
2		35(20.5%)	28(16.4%)	0.46
3		18(10.5%)	16(9.4%)	0.25
4		9(5.3%)	6(3.5%)	0.70
5 and above		5(2.9%)	2(1.2%)	0.75

The obstetric and gynecological characteristics i.e. parity, age at sexual debut and lifetime sexual partners were not significantly associated with an increased risk of any Pap smear abnormality (P value >0.05).

Table 8, correlation between contraceptive method, history of STI, HIV serostatus and any Pap smear abnormality.

Variable	Overall N=171	Any pap smear abnormality	Normal pap smear	Odds ratio 95% CI	P value
Contraceptive method used prior to the pregnancy					
None		43(25.1%)	37(21.6%)		
Natural methods		3(1.8%)	1(0.6%)		0.48
Barrier		4(2.3%)	1(0.6%)		0.32
Injectable progestin		15(8.8%)	10(5.8%)		0.82
Progestin implant		4(2.3%)	5(2.9%)		0.45
Combined pill		27(15.8%)	16(9.4%)		0.56
Intrauterine devices		2(1.2%)	0(0%)		0.22
History of STI					
Yes		9(5.3%)	7(4.1%)		0.68
No		90(52.6%)	63(36.8%)		
HIV serostatus					
Negative		89(52%)	68(39.8%)		
Positive		12(7%)	2(1.2%)	4.8(0.97-32.36)	0.027

The prior method of contraception and history of sexually transmitted infection were not statistically associated with abnormal pap smears, however mothers with positive HIV serostatus were more likely to have Pap smear abnormalities (odds ratio 4.8, p=0.027).

Table 9, Correlation between speculum examination findings and Pap smear abnormalities.

Variable	Overall N=171	Any pap smear abnormality	Normal pap smear	Odds ratio	P value
Vaginal discharge					
Normal		60(35.1%)	66(38.6%)		
Abnormal		36(21.1%)	4(2.3%)	9.75(3.07-34.4)	0.0000028
Gross examination of the cervix					
Normal		83(48.5%)	65(38%)		
Inflamed/cervicitis		12(7%)	4(2.3%)		0.16
Ulcer/mass		1(0.6%)	1(0.6%)		0.84

The presence of any abnormal vaginal discharge was associated with an increased risk of an abnormal pap smear result (odd ratio 9.75, P value=0.0000028). However, the gross appearance of the cervix was not statistically associated with pap smear abnormalities.

5.0 Discussion.

This study revealed a prevalence of 5.8% for squamous intraepithelial lesions (LSIL 3.5%, 2.3%HSIL). This is consistent with studies done in the US among pregnant women which documented prevalences of between 2% and 7% (4, 5). However the prevalence is lower than the rates of 12% and 14% documented by two separate studies conducted by Rukaria et al and Hugo De Vuyst et al among non pregnant women attending two family planning centers in Nairobi Kenya between the year 1998 and 2000 (10,11). The relatively lower prevalence could possibly be explained by the different populations studied.

This prevalence of 5.8% among women attending ANC in KNH may not be geneneralized to the rest of the country since KNH serves an urban and peri-urban population, and is also a referral hospital. Inferences can however be made and this high prevalence underlines the high burden of cervical dysplasia among pregnant women and underscores the importance of broadening antenatal care in our country to include early detection and management of cervical dysplasia. Failure to do so is a missed opportunity since 90% of pregnant women attend ANC at least once. Cervical cytology screening will improve reproductive heath care for women and will also have a significant impact on reducing morbidity and mortality from cervical cancer. This is further supported by the observation that 90% of this study's participants had not had previous screening for abnormal cervical cytology or cancer of the cervix . The 10.5% rate of screening recorded in this study is low compared to a rate of 22% found by Gichangi et al while

studying two groups of patients on follow up in KNH radiotherapy and obstetric/gynecology departments from the year 2000 to 2003 (58).

The Mean gestation age at enrolment (first booking visit) was 28 weeks. Most antenatal mothers (57.9%) came for their first antenatal visit in the 3rd trimester. This contrasts sharply with the situation in developed countries. For example, in the United States, 83.6% of the antenatal mothers began their ANC before 12 weeks gestation in the year 2004 (59).

This study recorded a high rate of inflammatory smears with no causative organism identified (31.6). Studies done elsewhere have recognized frequent inflammation as one of the factors that complicate sampling and analysis of cervical cytology in pregnancy (61). The 7% prevalence of bacterial vaginosis (diagnosed by the presence of clue cells) was clininically significant. Bacterial vaginosis has been associated with poor pregnancy outcomes.

The quality of pap smears in a pregnant population has traditionally been a concern. The rate of endocervical cell recovery in this study was 83%. This was consistent with a study done by Kruger et al in Philadelphia between May 2001 and May 2002 that found a rate of 82.1% and 90.5% in the liquid based smears and convectional smears respectfully (60). Only 2.9% of pap smears were inadequate for evaluation which is much lower than a rate 13.6% found in the study conducted by Kruger et al (60).

The prevalence of HIV infection in this study was 8.2% which is slightly higher than the 7.8% reported by Kenya AIDS indicator Survey conducted by NASCOP in

the year 2007 (62). HIV infection has been shown to increase the risk of cervical neoplasia by up to 5 times (16). This study showed a significant association between HIV infection and the presence of any cervical smear abnormality, with HIV sero-positive women 4.8 times more likely to have abnormal cervical smears. This emphasizes the urgency of introducing routine cervical cytology screening in our antenatal clinics and promoting counseling and testing for HIV.

Sexually transmitted infections, young age at sexual debut, multiple sexual partners, high parity and use of oral contraceptives have traditionally been associated with increased risk of abnormal pap smears. In this study, there was no significant association between abnormal cervical smears and parity, age of sexual debut, lifetime sexual partners, method of contraception and history of STIs. Presence of abnormal vaginal discharge was however noted to be significantly associated with abnormal cervical smears. This emphasizes the need to evaluate antenatal women with abnormal vaginal discharge and rule out reproductive tract infections.

Conclusion

The prevalence of abnormal cervical cytology was 5.8%.

Positive HIV sero-status and presence of abnormal vaginal discharge were noted to be associated with a higher risk of pap smear abnormalities.

The rate of previous screening for cervical cytology was low. Only 10.5% of the participants had had a pap smear done in the past.

Recommendation

Cervical cytology screening should be included in routine antenatal care in our hospitals. This would be in line with the WHO model on focused antenatal care which recommends cervical cytology screening on the first antenatal visit.

Antenatal care givers should actively promote and provide counseling and testing for HIV as this was observed to increase the risk of abnormal cervical cytology.

Antenatal mothers with vaginal discharge should be evaluated to rule out reproductive tract infections.

References:

1. National cervical cancer prevention program booklet, Action plan 2005-2009, Ministry of Health, Division of Reproductive Heath.
2. Deressa SS, Silverman DT, Young JL, et al. Cancer incidence and mortality trends among whites in the United States, 1947-1984. J Natl Cancer Inst 1987; 79:701-710.
3. Koss LG. The Papanicolaou test for cervical cancer detection; a triumph and a tragedy. JAMA 1989; 261:737-743.
4. Morimmura Y, Fujimori K, Soeda S, et al. Cervical cytology during pregnancy, comparison with non-pregnant women and management of pregnant women with abnormal cervical cytology. Fukushima J med 2002;48:27-37.
5. Douvier S, Filipuzzi L, Sagot P. Management of cervical intra-epithelial neoplasm during pregnancy. Gynecol Obstet Fertil 2003;31:851-5.
6. Pisani P, Bray, Parkin DM. Estimates of the worldwide prevalence of cancer for 25 sites in the adult population. Int J Cancer 2002; 97: 72-81.
7. Ndavi PM. Cervical cytology in a rural population. M Med Thesis. University of Nairobi. 1986; 417-450.
8. Kitavi JM. Cervical cytology in a rural area of Kenya. M Med Thesis. University of Nairobi; 1984: 386-417.

9. Chamia JM. The validity of Pap smear as a means of screening for cervical intraepithelial neoplasia (CIN) as compared to colposcopic and histologic findings. M.Med Thesis, University of Nairobi. 1986; 322-352.
10. M Temmerman, N Kidula, M tyndall, R Rukaria-Kaumbutho, L Muchiri, J O Ndinya-Achola. The burden of genital tract infections in a family planning clinic in Nairobi Kenya. Sex Trans inf 1998;74:202-204.
11. Hugo De Vuyst, et al. Distribution of Human Papilloma virus in a family planning population in Nairobi, Kenya. Sexually Transmitted diseases. Feb 2003 vol 30. NO.2, 137-142.
12. Morrison C, Catania F, Wakely P Jr, Nuovo GJ. Highly differentiated keratinizing squamous cell carcinoma of the cervix: locally aggressive tumor not associated with Human Papilloma virus or squamous intraepithelial lesion. Am J Pathol 2001; 25: 1310-5
13. Walboomers KM, Jacobs MV, Manos MM, et al. Human Papilloma virus is a necessary cause of invasive cervical cancer worldwide. J Pathol 1999; 189: 12-19.
14. Bosch FX, Manos MM, Munoz N, et al. Prevalence of Human Papilloma virus in cervical cancer, a worldwide perspective. Biological study on cervical cancer (IBSCC) study group. J Natl Cancer Institute 1995; 87:796-802.
15. Lungu O, Sun XW, Felix J, Richard RM, Silverstein S, Wright TC Jr. Relationship of human Papilloma virus type to grade of cervical intraepithelial neoplasia. JAMA 1992; 267:2493-6.

16. Frency A, Coutlee F, Franco E, Hankins C. Human Papilloma virus and HIV co-infection and the risk of neoplasia of the lower genital tract: a review of recent developments. CMA J 2003; 169:431-4.

17. Ault KA, Giuliano AR, Edwards RP, et al. A phase 1 study to evaluate a human Papilloma virus (HPV) vaccine. Vaccine 2004;22:3004-3007.

18. Harper DM, Franco EL, Wheeler CM, et al. Efficacy of a bivalent L1 virus-like particle vaccine in prevention of infection with human Papilloma virus types 16 and 18 in young women: a randomized controlled trial. Lancet 2004;364:1757-1765.

19. Harper DM, Franco EL, Wheeler CM, et al. Sustained efficacy of a bivalent L1 virus like particle vaccine against human Papilloma virus types 16 and 18:follow-up from a randomized controlled trial. Lancet 2006;367:1247-1255.

20. Koutsky LA, Ault KA, Wheeler CM, et al. A controlled trial of human Papilloma virus type 16 vaccine. N Engl J Med 2002;347:1645-1651.

21. Mao C, Koutsky LA, Ault KA, et al. Efficacy of human Papilloma virus-16 to prevent cervical intraepithelial neoplasia: a randomized controlled trial. Obstet Gynecol 2006;107:18-27.

22. Villa LL, Costa RI, Petta CA, et al. Prophylactic quadrivalent human papilloma virus (6, 11, 16 and 18) L1 virus like particle vaccine in young women: a randomized double blind placebo controlled multicentre phase 2 efficacy trial. Lancet Oncol 2005;6:271-278.

23. Pinto LA, Edwards J, Castle PE, et al. Cellular immune responses to human papilloma virus (HPV) 16 L1 in healthy volunteers immunized with recombinant HPV L1 virus like particles. J Infect Dis 2003;188:327-338.

24. Emeny RT, Wheeler CM, Jansen KU, et al. Priming of human Papilloma virus type 11-specific humoral and cellular immune responses in college aged women with a virus like particle vaccine. J Virol 2002;76:7832-7842.

25. US Food and Drug Administration. Product Approval information-Licensing Action: GARDASIL^{R} Questions and answers . Available at http://www.fda.gov/cber/products/hpvmer060806qa.htm.

26. McNealy SG Jr. New cervical cancer screening techniques. Am J Obstet Gynecol Oct 2003; 189: issue 4,supplement 1, pgs s40-s41.

27. Nuovo HM, Melnikov J, Howell LP. New tests for cervical cancer screening. Am Fam Physician 2001; 64: 780-786.

28. Bernstein SJ, Sanchez-Ramos I, Ndubis B. Liquid based cervical smear study and convectional Papanicolaou smears: a meta-analysis of prospective studies compairing cytologic diagnosis and sample adequacy. Am J Obstet and Gynecol 2001; 185: 308-317.

29. Hatch KD, Berek JS. Intraepithelial disease of the cervix, vagina and vulva in: Berek JS, ed. Novaks Gynecology 13th edition, Philadelphia. Lippincott Williams and Wilkins, 2002; 471-505.

30. Apgar BS, Zoschnik LB. Triage of the abnormal Pap smear. Prim Care. 1998; 25: 483-503.

31. Stillson T, Knight AL, Elswick RK Jr. The effectiveness and safety of two cervical cytology techniques during pregnancy. J Fam Practice 1997; 45: 159-163.

32. Paraiso MF, Brady K, Helmchen R, et al. Evaluation of the endocervical cyto brush and cervex brush in pregnant women. Obstet Gynecol. 1994: 84: 539-543.

33. Saslow, Debbie, Ronowiz, Carolyn N, et al. `American Cancer Society guidelines for early detection of cervical neoplasia and cancer'. CA Cancer J Clin 2002; 52: 342-362.

34. Michael CW, Esfahani FM. Pregnancy related changes; a retrospective review of 278 cervical smears. Diagn Cytopathol. 1997; 17: 99-107.

35. Londo R, Bjelland T, Girod C, et al. Prenatal and postpartum Pap smears: do we need both? Fam Pract Res J. 1994; 14: 359-367.

36. Lapolla JP, O'Neill C, Wetrich D. Colposcopic management of abnormal cervical cytology in pregnancy. J Reprod Med. 1988; 33: 301-306.

37. Hannigan EV. Cervical cancer in pregnancy. Clin Obstet Gynecol 1990; 33: 837-845.

38. Ostergard DR, Nieberg RK. Evaluation of abnormal cervical cytology during pregnancy with colposcopy. Am J Obstet Gynecol 1979; 134:756-758.

39. Economos K, Perez Verdiano N, Delke I, et al. Abnormal cervical cytology in pregnancy, a 17 year experience. Obstet Gynecol 1993; 81: 915-918.

40. Thomas C, Wright Jr, L Stewart et al;2006 ASCCP- sponsored consensus conference. 2006 consensus guidelines for the management of women with cervical cytological abnormalities. Am J Obstet Gynecol Oct 2007, 346-345.

41. Smith-Levitin M, Hernanez E, Anderson L, et al. Safety, efficacy, and cost of three cervical cytology sampling devices in prenatal clinic. J Reprod Me. 1996; 41: 749-753.

42. Wright TC Jr, Cox JT, Massad LS, et al. For the American Society for Colposcopy and Cervical Pathology. 2001 consensus guidelines for the management of women with cervical intraepithelial neoplasia. Am J Obstet Gynecol 2003; 189: 295-304.

43. Paraskevaidis E, Koliopoulos G, Kalantaridous s, Et al. Management and evaluation of cervical intraepithelial neoplasia during pregnancy and post partum. Eur J Obstet Gynecol Reprod Biol 2002; 104: 67-69.

44. Siddiqui G, KurZel RB, Lampley EC, et al. Cervical dysplasia in pregnancy. Progression versus regression postpartum. Int J Fetil Womens Med. 2001; 46: 278-280.

45. Kiguchi K, Bibbo M, Hasegawa T, et al. Dysplasia during pregnancy: a cytological follow up study. J Reprod Med 1981; 26: 66-72.

46. Ueki M, Ueda M, Kumagai K, et al. Cervical cytology and conservative management of cervical dysplasia during pregnancy. Int J Gynecol Pathol. 1995; 14: 63-69.

47. Yoonesi M, Wieckowska W, Mariniello D, et al. cervical intraepithelial neoplasia in pregnancy. Int J Obstet Gynecol. 1982; 20: 111-118.

48. Ahdoot , Van nostrand KM, Nguyen NJ, et al. The effect of route of delivery on regression of abnormal cervical cytology findings in the postpartum period. Am J Obstet Gynecol; 1998; 178: 1116-1120.

49. Woodrow N, Permezel M, Butterfield L, et al. Abnormal cervical cytology in pregnancy: experience of 811 cases. Aust NZJ Obstet Gynecol. 1998; 38: 161-165.

50. Averette HE, Nasser N, Yankow SL, et al. Cervical conization in pregnancy. Analysis of 180 operations. Am J Obstet Gynecol. 1970; 106: 543-549.

51. Berman ML, Disaia PJ< Brewster WR. Pelvic malignancies, gestational trophoblastic neoplasia, non-pelvic malignancies. In: Creasy RK, Resnik R, eds. Maternal fetal medicine. 4th ed. Philadelphia-PA: WB Saunders; 1999: 1128-1132.

52. Goldberg GL, Altaras MM, Block B. Cone cerclage in pregnancy. Obstet Gynecol 1991; 77: 315-317.

53. Dunn TS, Ginsburg V, Wolf D. Loop-cone cerclage in pregnancy: a 5 year review. Gynecol oncol. 2003; 90:577-580.

54. Robinson WR, Webbs, Tirpack S, et al. Management of cervical intraepithelial neoplasia with loop excision. Gynecol Oncol. 1997; 64: 153-155.

55. Goff BA, Paley PJ, Koh WJ, et al. Cancer in the pregnant patient. In: Hoskins WJ, Perez CA, Young RC eds. Principles and practice of Gynecologic , 3rd ed. Philadelphia PA. Lippincott Williams and Wilkins; 2000: 501-528.

56. Creasman WT. Cancer in pregnancy. In: Gilstrap LC III, Cunningham FG, Vandorsten JP, eds. Operative Obstetrics. 2nd ed. New York, NY: McGraw-Hill; 2002: 424-426.

57. Disaia PJ, Creasman WT, eds. Clinical Gynecologic oncology. 5th ed. St Louis, MO: Mosby; 1997: 446-452.

58. P. Gichangi etal. Knowledge and practice about cervical cancer and pap smear testing among patients in Kenyatta National Hosp, Nairobi, Kenya. Int J Gynecol Cancer 2003:13, 827-833.

59. Brady E, Hamilton, Ariald M, et al. Annual Summary of Vital Statistics 2005. *Pediatrics* 2007; 119:345-360.

60. Kruger et al. Randomized pilot study comparing rates of endocervical cell recovery between convectional and liquid based cytology in a pregnant population. J lower Gen Tract disease 2003:vol7; Num 2, 101-103.

61. Pisharodi LR, Jovanoska s. Spectrum of cytologic changes in pregnancy. A review 100 abnormal cervicovaginal smears, with emphasis on diagnostic pitfalls. Acta cytol 1995; 39:905-8.

62. Kenya AIDS indicator survey conducted in women by NASCOP and National AIDS Control Council in the year 2007.

PATIENT INFORMATION AND CONSENT FORM.

Study to determine the prevalence of abnormal cervical smears among women attending antenatal clinic at Kenyatta National Hospital.

Investigator: Dr Samuel Wachira Ndungu, Masters of medicine student, Department of Obstetrics and Gynecology, University of Nairobi.

Supervisors

Dr Zahida Qureshi, Consultant Obstetrician and Gynecologist, Senior Lecturer, University of Nairobi.

Dr Harrisson Tamoo, Obstetrician and Gynecologist, Kenyatta National Hospital.

KNH Ethical Review commitee Chairperson, Professor K.M Bhatt, Phone number 0202726300.

Researcher's statement

I am asking you to participate in a research study. The purpose of this form is to give you information about the study on abnormalities of cells in the birth canal, specifically the cervix (the opening of the womb), among women attending antenatal clinic at Kenyatta National Hospital. This information will help you decide whether to be in the study or not. Please read the form carefully. You may ask questions about the purpose of the research, what we would ask you to do, the possible risks and benefits, your rights as a volunteer and anything else about the research. When we have answered all questions, you can decide to participate in the study or not. If you wish, we will give you a copy of this form for your records.

Purpose and benefits

The purpose of this study is to document the prevalence of abnormal cells in the birth canal among pregnant women attending clinic at Kenyatta National Hospital. Some of the abnormalities can predispose to cancer of the cervix (opening of the birth canal). Most women with abnormal cells can be treated successfully if discovered early. This study will be very useful in the prevention of cancer of the cervix, the commonest cancer in our country. In addition , you will be given your pap test results and you will be advised on follow up.

Procedure

I and my research assistant will obtain information about you using a questionnaire. We will subsequently examine your birth canal and collect a sample from the cervix. A speculum (an instrument used to part the opening of the birth canal) will be inserted to allow visualization of the cervix and collection of the sample. The sample will then be taken to the laboratory for assessment. As you participate in the study, you will also receive the standard antenatal care at Kenyatta National Hospital.

Risks, Stress or Discomfort.

- Completing the questionnaire and examination of your birth canal will take about 15 minutes of your time.
- Examination of your birth canal with the aid of a speculum could cause discomfort.

- In the event that your test results are abnormal, you will be referred to a specialist clinic (gynecology) for treatment. Depending on the severity or stage of the pap test abnormality, some of the treatment methods could cause a miscarriage or preterm delivery (birth of the baby before the due date).

Confidentiality

All the information obtained from you will be treated with utmost confidentiality. Your name will not appear on the questionnaire or the lab request form. A study number will be used instead. Your pap test results will be filed with your antenatal clinical records.

You may choose to withdraw from the study, refuse to answer questions or decline the pap test. Your decision will not affect your antenatal care at KNH.

Investigators signature..... Date.....

Subject's statement

I voluntarily agree to participate in the study on prevalence of abnormal cells in the birth canal of pregnant women at Kenyatta National Hospital clinic 18. I understand that participation in the study does not entail financial benefit. I have been informed that information obtained will be treated with utmost confidentiality and my treatment will not be compromised if I decline participation or withdraw from the study.

I have had a chance to ask questions. If I have questions later, I can ask the researcher. If I have questions later about my rights as a research subject, I can call the ethical review committee at Kenyatta National Hospital on phone number 020726300.

Subject's signature..... Date.....

OR

Subject's left thumb print..... Date.....

Subject's name.....

Prevalence of abnormal cervical cytology smears among women attending ANC at Kenyatta National Hospital.

Pap smear Request Form

Serial Number: [ ]

1.Age (years)

2.Gestation (weeks)

3.Gross appearance of the cervix.....

4.Other clinical details.....

.....

5.Date of Specimen Collection:.....

6. Report. (Please tick relevant box)

6.1 Specimen Adequacy

☐ Satisfactory for evaluation

☐ Specimen evaluated but limited by

- Excess blood
- Lack of endocervical cells
- Others (specify).....

.....

☐ Unsatisfactory for evaluation (not interpretable)

Reason.....

6.2 Results.

- [ ] Normal

6.3 Organisms

- [ ] Trichomonas vaginalis
- [ ] Fungal organisms consistent with Candida spp.
- [ ] Shift in flora suggestive of bacteria vaginosis
- [ ] Bacteria morphologically consistent with Actinomyces
- [ ] Cellular changes associated with herpes simplex virus

6.4 Squamous cell abnormalities

- [ ] Atypical squamous cells of undetermined significance (ASCUS).
- [ ] Atypical squamous cells cannot exclude HSIL (ASC-H)
- [ ] Low-grade squamous intraepithelial lesion (LSIL)
- [ ] High-grade squamous intraepithelial lesion (HSIL)
- [ ] Squamous cell carcinoma.

6.5 Glandular cell abnormalities.

- ☐ A typical endocervical cells (NOS or specify).
- ☐ A typical endometrial cells (NOS or specify)
- ☐ Atypical endocervical cells favour neoplasia.
- ☐ Atypical endometrial cells favour neoplasia.
- ☐ Adenocarcinoma insitu.
- ☐ Invasive adenocarcinoma

6.6 Remarks OR other abnormalities

.....

.....

Name of cytologist Reporting.....

Signature.....Date.....

Quality Control Cytologist.....

Signature.....Date.....

Prevalence of abnormal cervical cytology among women attending ANC at Kenyatta National Hospital.

Questionnaire

1. Age of participants (years). [ ]
2. Gestation by dates (weeks) [ ]
3. Marital status
   a. Single
   b. Married
   c. Divorced/separated
   d. Widowed
4. Level of education
   a. No formal education
   b. Primary
   c. Secondary
   d. University/college
5. Occupation
   a. Unemployed
   b. Self employed
   c. Employed (casual)
   d. Skilled employment (professional)

6. Number of births

a. none

b. I

c. 2

d. 3

e. 4

f. >4

7. Number of abortions

a. none

b. 1

c. 2

d. 3

e. >3

8. Age at first sexual intercourse.

a. 0-9

b. 10-14

c. 15-19

d. 20-24

e. 25-29

f. 30+

9. Number of lifetime sexual partners.

   a. 1

   b. 2

   c. 3

   d. 4

   e. 5 and above

10. Age at first pregnancy

   a. 10-19

   b. 20-29

   c. 30-39

   d. 40-49

11. Contraceptive method used prior to the pregnancy

   a. None (go to 13)

   b. Natural methods

   c. Barrier methods

   d. Injectable progestin

   e. Progestin implants

   f. Combined oral pill

   g. Intrauterine devices

12. Duration of contraception

a. < 1 year

b. 1-3 years

c. >3 years.

13. History of sexually transmitted infections.

a. Yes

b. No

15. Cigarette smoking

a. No

b. Yes

16. HIV status (Record from antenatal profile)

a. Negative

b. Positive

17. Has Pap smear been done before?

a. Yes (go to 18)

b. No

18. What was your Pap test result?

a. Normal

b. Abnormal (specify)

c. I do not know.

Speculum examination

19. Vaginal discharge

a. Normal

b. Abnormal (specify)

20. Gross examination of the cervix

a. Normal/healthy

b. Inflammed/cervicitis

c. Ulcer/mass

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Ref: KNH-ERC/ 01/ 4996

5th December 2007

Dr. Samuel Wachira Ndungu
Dept. of Obs/Gynae
School of Medicine
University of Nairobi

Dear Dr. Wachira

RESEARCH PROPOSAL: "PREVALENCE OF ABNORMAL CERVICAL CYTOLOGY SMEARS AMONG WOMEN ATTENDING ANENATALCLINIC IN K.N.H" (P262/09/2007)

This is to inform you that the Kenyatta National Hospital Ethics and Research Committee has reviewed and approved your revised research proposal for the period 5th December 2007 – 4th December 2008.

You will be required to request for a renewal of the approval if you intend to continue with the study beyond the deadline given. Clearance for export of biological specimen must also be obtained from KNH-ERC for each batch.

On behalf of the Committee, I wish you fruitful research and look forward to receiving a summary of the research findings upon completion of the study.

This information will form part of database that will be consulted in future when processing related research study so as to minimize chances of study duplication.

Yours sincerely

PROF C. KIGONDU
AG. SECRETARY, KNH-ERC

c.c. Prof. K.M. Bhatt, Chairperson, KNH-ERC
The Deputy Director CS, KNH
The Dean, School of Medicine, UON
The Chairman, Dept. of Obs/Gynae, UON
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