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**HUMAN PAPILLOMA VIRUS AND HUMAN IMMUNODEFICIENCY VIRUS
INFECTIONS AMONG MEN WHO HAVE SEX WITH MEN IN MACHAKOS
COUNTY, KENYA**

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P150/22958/2011**

**1
A RESEARCH PROPOSAL SUBMITTED IN PARTIAL FULFILMENT OF THE
REQUIREMENTS FOR THE AWARD OF THE DEGREE OF MASTER
DEGREE IN INFECTIOUS DISEASES (CLINICAL VIROLOGY) IN THE
SCHOOL OF HEALTH SCIENCE OF KENYATTA UNIVERSITY**

January, 2024

DECLARATION

This proposal is my original work and has not been presented for the award or degree or any other award in any university.

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

Ndolo Elizabeth Munee

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1
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ABBREVIATIONS AND ACRONYMS

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GbMSM – gay, bisexual, and men who have sex with men

HBM – Health Belief Model

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HIV – Human immunodeficiency virus

HPV – Human papilloma virus

MSM – Men who have sex with men

MoH – Ministry of Health

SEM – Social Ecological Model

WHO – World Health Organization

PLHIV-- People living with HIV

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NACOSTI--National Commission for Science, Technology and Innovation

AGYW -- Adolescent girls and young women (AGYW)

STI – Sexually transmitted diseases

AIDs –Acquired immunodeficiency syndrome

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DNA--Deoxyribonucleic acid

EDTA—Ethylenediamine tetra acetic acid

VCT—Voluntary HIV counseling testing

ABSTRACT

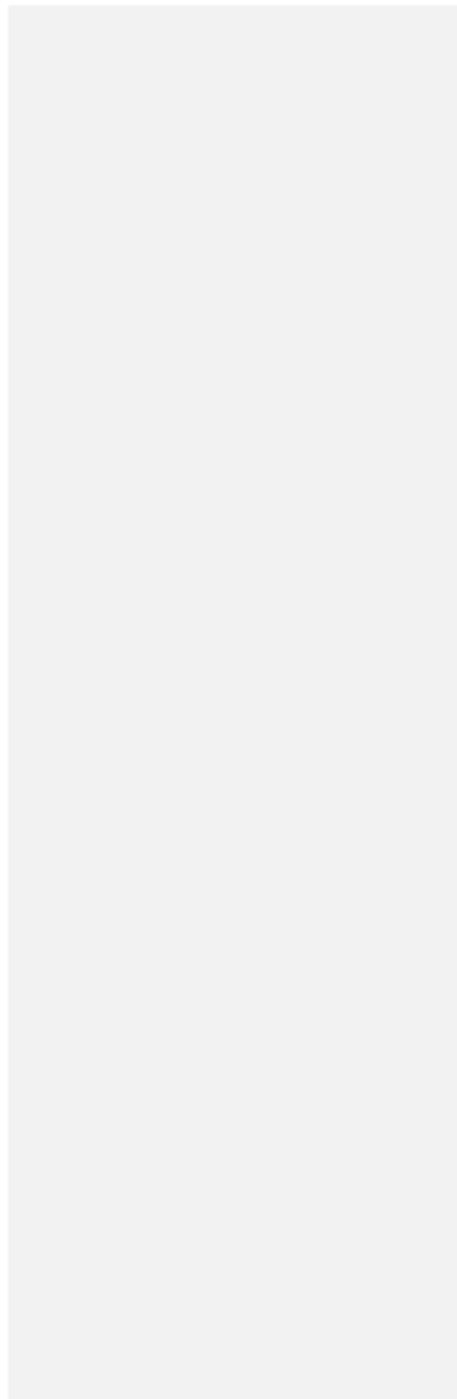
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This study aims to comprehensively understand the epidemiology of Human Papillomavirus (HPV) and HPV/Human Immunodeficiency Virus (HIV) coinfection among men who have sex with men (MSM) in Machakos County, Kenya. As both HPV and HIV coinfection poses significant health challenges, particularly in the MSM community, a descriptive cross-sectional study will be conducted in Dice Clinics in Machakos County. The research will focus on five specific objectives. First, to determine the available genotype of HPV among MSM at Dice Clinics; second, to establish the prevalence of HPV among this population; third, to assess the prevalence of anogenital cancer among MSM; fourth, to determine the prevalence of HPV/HIV coinfections and fifth, determine risks factors associated with HPV among MSM at Dice Clinics in Machakos County, Kenya. Kenya, in its pursuit of Universal Health Care, lacks sufficient data on the epidemiology of HPV/HIV coinfection among MSM, especially in periurban areas like Machakos County. The study will employ random sampling, aiming to recruit 780 MSM attending Dice Clinics over three months. Structured questionnaires will capture socio-demographic and risk-related data, and HIV and HPV testing done. The analysis, using SPSS statistical software, will involve data cleaning, processing, and a thorough examination of variables. Chi-square tests will compare proportion values, and correlation coefficients will explore relationships between study variables. Ethical considerations are paramount, and approval will be sought from Kenyatta University Ethical Review Committee and the National Commission for Science, Technology, and Innovation. Informed consent will be obtained from participants, ensuring respect for their autonomy and privacy throughout the study. This research is pivotal for filling the information gap on HPV/HIV coinfection among MSM in Kenya, contributing to better healthcare strategies and ultimately enhancing the inclusivity of Universal Health Care efforts in the country.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background of the study

Human Papillomavirus (HPV) and Human Immunodeficiency Virus (HIV) have been identified as two of the most commonly infections related to sexual activity among different populations, drawing a lot of research interest into their potential, especially coinfection trends among select study groups. Virus-related cancer cases have been tallied to account for up to 15% of all cases globally. 50,000 cervical cancer associated deaths annually in the sub-Saharan region, this rate was likely to rise even further in coming Years due to coinfection with HIV. In Kenya cancer cases have been on the rise in recent years, and being a country that embraces diverse sexual orientations and preferences, the country has become a major hub for data on the epidemiology of a number of cases in Africa and the world at large. Nairobi, Nakuru and Kiambu are the three counties in Kenya with the highest number of cancer cases, with Machakos only accounting for 465 cases of the 42166 cancer cases in Kenya in 2022. The county, therefore, has been viewed as a strategic study location as it would help paint a picture that is representative of the entire nation due to its multicultural diversity.

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Both HPV and HIV pose grave public health challenges, especially where the rate of infection is high while the coverage of vaccines is low (Chambers, et al., 2022). It has been found that people living with HIV (PLHIV) are generally at a high risk of developing HPV-related cancers, with cervical cancer as a designated AIDS-defining condition in different populations all across the world (Lekoane, Kuupiel, Mashamba-Thompson, & Ginindza,

Commented [AMM1]: Review the global, regional and Kenya burden of HIV, HPV and co-infections in general population and MSM

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2020). World Health Organization (WHO) estimates that 625 600 women and 69 400 men get an HPV-related cancer each year, with prevalence being mostly in low- and middle-income countries, which experience three times higher occurrence compared to high income countries (Kangethe, et al., 2023).

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Several studies have explored the prevalence and risk factors of Human Papillomavirus (HPV) and HIV coinfections among men who have sex with men (MSM) in diverse contexts. Remis et al. (2016) conducted a study in Toronto, focusing on 442 MSM, of which 294 were living with HIV, and revealed that MSM with HIV had a higher predisposition to HPV coinfection (67.6%) compared to those without HIV (51.7%). Risk factors included age, number of sexual partners, and injected drug use. Rodríguez-Álvarez et al. (2018) in Spain, identified race, circumcision, number of female partners, and engaging in sex with men as major factors influencing HPV infection among men. Lekoane et al. (2020) addressed the interplay of HIV and HPV-related cancers in sub-Saharan Africa, emphasizing a literature gap in HPV-related cancers among people living with HIV in many SSA countries. They reported poor participation among men in data collection. Müller (2016) in Cape Town, South Africa, found a high prevalence of HPV (72.8%) among 200 MSM, with 88 of them being HIV positive. Risk factors for HPV infection included having sex with men only, engaging in group sex, living with HIV, and practicing receptive anal intercourse.

Gay, bisexual, and men who have sex with men (gbMSM), stand at great risk of infection with both HPV and HIV, mainly owing to a number of factors that hinder their access to

preventive and curative healthcare services in Kenya, and indeed, globally. Some of the hindering factors are self-inflicted while others are from external sources. It has been established, for example, that Gardasil human papillomavirus (HPV) vaccine, which could be useful to this target group is not readily available to them, as it only freely available to adolescent girls and young women (AGYW) (Joint Committee on Vaccination and Immunisation, 2015). With an already overburdened healthy system that does not have adequate supply of HPV vaccines to cater for the primary target group which constitutes the adolescent girls and young women, MSM are left out of free and adequate access of the vaccines, greatly exposing them to the danger of coinfection between HPV and HIV (Remis, et al., 2016). Additionally, social stigma associated with the identification as men who have sex with men also makes a number of men fail to acquire the vaccines from medical facilities, which creates a bedding ground for coinfection between HPV and HIV (Baseman & Koutsky, 2005). The use of PrEP and condoms among MSM, while generally reported to reduce the risk of co-infection is faced with several hindrances like access to the condoms as well as failure to use them, owing to personal preferences among MS (Luchters, et al., 2010).

1.2 Statement of the problem

Both HPV and HIV present significant public health challenges, particularly in areas with high infection rates and low vaccine coverage. Individuals with HIV face an elevated risk of developing HPV-related cancers, including cervical cancer, recognized as an AIDS-defining condition. Notably, men who have sex with men (MSM) have emerged as a specific group under scrutiny in recent research endeavors (Kangethe et al., 2023).

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Kenya has placed keen emphasis on Universal Health Care (UHC) to all, including individuals considered to be part of minority groups. However, information showing the epidemiology of HPV/HIV coinfection among men who have sex with men in Kenyan periurban areas is limited, which would be crucial in helping in policy development to reduce risk of exposure to HPV and HIV to the general population, considering that some of the MSM also sleep with women.. Therefore, this study will determine the epidemiology of HPV, HIV and their coinfection among men who have sex with men (MSM) in Machakos County, Kenya. Additionally, the risk factors for acquisition of HPV and HIV among the study group will be elucidated to provide insights into the complex dynamics of these infections for targeted prevention and control interventions.

1.3 Justification

Despite the Ministry of Health (MoH) commitment to quality medical care access to all Kenyans, there still exists a huge barrier in regards to access and consumption of such services by members of minority groups such as men who have sex with men. Even where such service are accessible, it has not been easy to take note of the patterns and spread of some pertinent health issues, such as the coinfection of HPV and HIV, which has stood in the way of understanding critical information that would help enhance the penetrance and effectiveness of medical interventions among men who have sex with men.

In Africa and particularly in Kenya, the epidemiology and the risk factors associated with HPV/ HIV coinfection among men who have sex with men are poorly investigated or remain largely uninvestigated in several areas. In Machakos County, HPV is not a routine

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Commented [WU6R5]: If not provided with proper medical care, they can hinder eradication of disease considering that some of them still have sex with women, meaning they could facilitate the spread of HPV and HIV.

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test among men who have sex with men, and especially those who have already been infected with HIV. As can be noted, there has been a number of studies conducted on the prevalence of HPV among MSM have been done in Kenya but no publicly available study has specialized on conducting this research in periurban areas in Kenya. This study, therefore, will seek to understand the occurrence of HPV/HIV coinfection and the prevalent HPV genotypes among MSM who attend Dice Clinics in Machakos County.

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1.4 Research questions

- i. What is the prevalence of HPV infections and HPV genotypes among MSM in Machakos County?
- ii. What is the prevalence of HIV and HPV coinfections among MSM at Dice Clinics in Machakos County?
- iii. What is the prevalence of anogenital cancer among MSM infected with high-risk HPV genotypes in Machakos County?
- iv. What are the risks factors associated with HPV and HIV infections among MSM in Machakos County?

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1.5 Study objectives

1.5.1 General objective

To determine the epidemiology of HPV and HIV infections among men who have sex with men (MSM) in Machakos County, Kenya.

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1.5.2 Specific objectives

- i. To determine prevalence of HPV infections and HPV genotypes among MSM in Machakos County.
- ii. To determine the prevalence of HIV and HPV coinfections among MSM at Dice Clinics in Machakos County.
- iii. To determine the prevalence of anogenital cancer among MSM infected with high-risk HPV genotypes in Machakos County.
- iv. To elucidate the risks factors associated with HPV and HIV infections among MSM in Machakos County.

1.6 Significance and anticipated output of the study

The study serves as a comprehensive resource with practical implications for various stakeholders, including practitioners, academic researchers, and those shaping the regulatory landscape in Kenya's public health and medical sectors. Research examining the epidemiology of HPV, HPV/HIV coinfection among men who have sex with men, specific in a periurban area like Machakos County in Kenya, presents policymakers and the government with valuable insights. The findings of this research would be essential in shaping the regulatory frameworks and policies that affect medical care for diverse groups of target groups within the population, and especially minority groups, such as men who have sex with men. This can touch on issues such as access to vaccines and related medical services. For practitioners within the medical fraternity, this study offers insights into the specific impacts of the HPV, HPV/HIV coinfection among MSM. This would enable them to adapt strategies that enhance service delivery and especially in the diagnosis and treatment in their professional endeavors. Scholars engaged in the medical sector would

benefit from the empirical data and case study approach employed in this research. The study would deepen their understanding of existing nuanced correlation amongst HPV and HIV infection among MSM and associated risk factors, contributing to academic discourse on this subject. The research findings will provide a significant contribution to the broader knowledge on public health. The findings offer refined perspectives on this topic, thereby influencing national public health, especially being that the country is facing pressure to decriminalize different sexual orientation and preferences which includes legal and social acceptance of men who have sex with men.

CHAPTER TWO**2.0 LITERATURE REVIEW****2.1 HPV/HIV coinfection**

HPV has been reported by several studies to have a high degree of coinfection with HIV among different target groups. This pattern transcends geographical, sexual orientation and any other barriers. Studies conducted from a global, regional and even national level show consistency in this pattern, which has raised the need to conduct more in depth research among different targets groups defined by several factors such as men who have sex with men, as well as women who work as sex workers in different geographical regions. Coinfection between Human Papillomavirus (HPV) and Human Immunodeficiency Virus (HIV) presents a complex and multifaceted challenge, particularly among men who have sex with men (MSM). MSM, a demographic disproportionately affected by these infections, encounter heightened risks due to the interconnected nature of HPV and HIV transmission. HPV, a group of sexually transmitted viruses, comprises numerous strains with varying degrees of risk.

Thirteen high-risk HPV strains are identified as carcinogenic, with HPV16 being more prevalent in anal cancer among MSM compared to cervical cancer (Smith, et al., 2010). This elevated risk underscores the importance of exploring the dynamics of coinfection within this specific demographic. The relationship between HPV and HIV is bidirectional. Individuals living with HIV face an increased susceptibility to HPV-related cancers. The compromised immune system in HIV-positive individuals allows persistent HPV infections, leading to a higher likelihood of developing cancers, particularly in anogenital

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areas. Conversely, having HPV may facilitate HIV acquisition, as genital warts caused by certain HPV strains create entry points for the virus (Lorway, et al., 2022).

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2.2 Risk factors of HPV/HIV coinfection among MSM

Several risk factors contribute to the heightened prevalence of HPV/HIV coinfection among MSM. Unprotected anal sex, a common practice among this demographic, amplifies the risk of both infections. Immunocompromised individuals, often a characteristic of those with HIV, struggle to clear HPV infections, leading to persistence and potential malignancy. The intricacies of sexual networks within the MSM community further complicate prevention efforts, as the interconnectedness of partners increases the likelihood of viral transmission. Substance use, particularly stimulant drugs, is another risk factor. Substance use can influence sexual behaviors, leading to riskier practices and increased vulnerability to infections (Grace, et al., 2018). Moreover, the shared use of drug paraphernalia may contribute to the transmission of both HIV and HPV.

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Efforts to address HPV/HIV coinfection among MSM must consider these risk factors comprehensively. Tailored interventions should prioritize safe sexual practices, regular screenings, and vaccination against preventable HPV strains (Lorway, et al., 2022). Holistic healthcare strategies that account for the interplay between immunosuppression, sexual behaviors, and substance use are crucial in mitigating the impact of coinfection within the MSM community.

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2.3 Genotypes of HPV among MSM

A comparison between the rate of infection of HPV between ²men who have sex with men shows that those ¹²who already have HIV are more susceptible to being infected with HPV than those who are not already infected with HIV. It was also found ⁵⁴that men who have sex with men and had been infected with HIV were five percent ⁴⁷more likely to suffer from anal cancer in comparison to HIV-negative ⁵men who have sex with men. Of the numerous types of HPV transmitted through sexual contact, primarily affecting the anogenital areas, 13 high-risk HPV strains (HPV16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 68) are recognized as carcinogenic, with HPV16 being more commonly associated with anal cancer related to HPV than with cervical cancer ⁵(Oo, et al., 2023). While HPV vaccination and the prevention and control of HIV infection are two primary prevention strategies for mitigating the coinfection rates, it has been found that there are prevailing risk factors that ³²prevent men who have sex with men from accessing proper medical treatment that would work to reduce this coinfection ¹⁹(Grace, et al., 2018).

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2.4 Treatment

There remains no definitive cure for HPV infections, with the focus primarily on symptom management and prevention. Treatment modalities address specific manifestations, such as topical medications and procedures for genital warts, and monitoring or intervention for precancerous lesions (Rodríguez-Álvarez *et al.*, 2018). Vaccines play a pivotal role in preventing Human Papillomavirus (HPV) infections. Currently available vaccines, such as Gardasil and Cervarix, target high-risk HPV types associated with cervical and other cancers, ¹³as well as low-risk types causing genital warts (Remis *et al.*, 2016). While

traditionally recommended for females, these vaccines are also crucial for men who have sex with men, offering protection against related cancers and reducing transmission. In recent years, efforts have expanded to increase vaccine accessibility and uptake across diverse populations, emphasizing the importance of gender-neutral vaccination strategies.

2.5 Prevalence and known risk factors across the world

Remis *et al.*, (2016) while studying the prevalence of Sexually Transmitted Viral and Bacterial Infections in HIV-Positive and HIV-Negative Men Who Have Sex with Men in Toronto studied 442 MSM, where 294 were living with HIV and 148 were not. The study found that men who have sex with men and living with HIV were more predisposed to HPV coinfection compared to those who did not have HIV, at 67.6% versus 51.7% respectively. Some of the risk factors identified for coinfection included the age and number of sexual partners as well as use of injected drugs. Rodríguez-Álvarez *et al.*, (2018) conducted a study on the prevalence and risk factors of Human Papillomavirus in male patients in Spain. An analysis of existing literature revealed that race, circumcision, number of female partners and having sex with men also play a major factor in influencing HPV infection among men.

Lekoane *et al.*, (2020) in a study on the interplay of HIV and human papillomavirus-related cancers in sub-Saharan Africa reported that there exists a literature gap on the HPV-related cancers among people living with HIV individuals in many countries within SSA. It was further reports that very few studies focused on urban areas and none had been conducted in rural areas. In collecting the data, the researchers encountered poor participation among

men. On the other hand, Müller (2016) ³ investigated the prevalence of human papillomavirus (HPV) infection and associated behavioral risk factors in men-who-have-sex-with-men (MSM) attending a clinical service in Cape Town, South Africa. The study found that 72.8% of the 200 participants were infected with HPV, with 88 of the 200 being HIV positive. 91.8% of the MSM living with HIV had an anal HPV infection, while 81.2% ³¹ had anal HR-HPV and 85.9 % had multiple anal HPV types. The only independently associated risk factors for HPV infection in this study were, having sex with men only, ⁶¹ engaging in group sex in lifetime, living with HIV and practicing receptive anal intercourse. ³

Oo *et al.*, reported that Human papillomavirus (HPV) infection had a profound affiliation with anal cancers, predominantly in gay, bisexual, and men who have sex with men (gbMSM), largely owing to their susceptibility to HIV infection. ⁵ Of the 115 participants, 61 of the MSM were HIV negative while 51 were HIV positive. After stratifying the results by HIV status, the HPV prevalence was found to be 84.3% for gbMSM living with HIV ³² and 24.6% for gbMSM without HIV. ⁵ The study found that there were certain risk factors ²³ that seemed to define gbMSM who were infected with both HPV and HIV. One of the most predominant factors included the fact that it seemed a greater number of those who had the coinfection were also married to a woman. ¹⁰ Kang'ethe *et al.*, (2023) found that women living with HIV on antiretroviral therapy at the Kenyatta National Hospital in Kenya exhibited a higher degree of susceptibility to coinfection with HPV. The study reported that while there are about 50,000 cervical cancer associated deaths annually in the sub-Saharan region, this rate was likely to rise even further in coming Years due to coinfection

with HIV. Of the 647 respondents, it was noted that there were several risk factors that promoted the HPV/HIV coinfection, including having several sex partners, alcohol consumption as well as whether or not their sexual partner was circumcised. The study found that only 1.1% of the respondents were vaccinated against HPV, many of the unvaccinated being part of women in low and middle income households.

In different study by Smith *et al.*, (2010) it was found that circumcision was a crucial determinant of the epidemiology of HPV infections among men, providing a major source of information as to the coinfection between HPV and HIV. Out of the 2705 participating men, 51.1% were found to have HPV, and although the study did not test for whether these uncircumcised men had sex with men, it provided a groundbreaking source of information to related circumcision to HPV/ HIV coinfection among MSM. In another study to test for increased risk of HIV acquisition among Kenyan men with Human Papillomavirus infection, it was found that of the 2168 uncircumcised men living with HPV, 5.8% of them were also coinfecting with HIV, showing that circumcision was also a risk factor exposing men to coinfection with HPV and HIV (Smith *et al.*, 2010).

Despite the critical need for more studies, it is evident that there has not been adequate studies to investigate the coinfection between HPV and HIV, and particularly in periurban areas, which is the premise upon which this research will be based.

1 CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area

The study will be carried out in Machakos County in Kenya. Machakos is a budding periurban area which has been attracting a large number of middle class working-age individuals, and has been seen recent attention due its strategic economic development that has attracted people of different inclinations in terms of sexual preference. Machakos County's administrative headquarters are located in Machakos town, and the county has a population of 1,421,932 as of 2019, out of whom 150,041 live in the metropolitan areas. Machakos county neighbors Nairobi and Kiambu counties on its west side, Embu on its north side, Kitui on its east side, and Makueni on its south side (Munguti, 2022). Nairobi, Nakuru and Kiambu are the three counties in Kenya with the highest number of cancer cases, with Machakos only accounting for 465 cases of the 42166 cancer cases in Kenya in 2022. Machakos is a rapidly developing periurban area that has been reported to have a higher number of MSM seeking medical treatment in medical facilities, as a result of which Machakos presents a favorable research area (Kangethe et al., 2023). The presence of Dice Clinics which records up to 13 MSM seeking HPV or HIV related medical care also adds to the suitability of Machakos as the area of study. The clinics offer rapid HIV test as well as HPV tests through anogenital swabs, counselling and related care for MSM in Machakos County.

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Commented [AMM7]: Why are you proposing this study in Machakos considering the biggest problem is in Nairobi, Nakuru and Kiambu. This should come out clearly
-Again, the study area should contain the DICE centres location, the number of clients received per day or month, the kind of routine services provided.....

Commented [WU8R7]: Machakos is a rapidly developing periurban area that has been reported to have a higher number of MSM seeking medical treatment in medical facilities, as a result of which Machakos presents a favorable research area (Kangethe et al., 2023). The presence of Dice Clinics which records up to 13 MSM seeking HPV or HIV related medical care also adds to the suitability of Machakos as the area of study. The clinics offer rapid HIV test as well as HPV tests through anogenital swabs, counselling and related care for MSM in Machakos County.

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According to the ²¹ National AIDS and STI Control Council (NASCOP) of the Ministry of Health which carried out a research in collaboration with the University of Manitoba, it was estimated that there are about 19,175 MSM in Kenya, with the four counties having highest population of this community being Nairobi at 11,000, Kiambu at 3635, Kisumu at 1567 and Mombasa at 1469. Although less than 1000 MSM were reported in Machakos County, it was observed as an area with rising numbers of MSM in the report, making it a suitable study area. The MSM community was found to have a HIV prevalence of 18.2% ²⁷ which was considered three times higher than that of the general population (Oo, et al., 2023). The county, therefore, has been viewed as a strategic study location as it would help paint a picture that is representative of the entire nation due to its multicultural diversity.

Commented [AMM9]: The question is: where is Machakos in this data? See above comment

Commented [WU10R9]: Although less than 1000 MSM were reported in Machakos County, it was observed as an area with rising numbers of MSM in the report, making it a suitable study area.

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3.2 Study Population

The study population will be ² men who have sex with men (MSM) attending DICE centers in Machakos County.

3.2.1 Inclusion criteria

⁶² Both HIV negative and HIV positive gays and bisexuals.

3.2.2 Exclusion criteria

³ Men who have sex with men not consenting to the study participation.

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3.3 Study design

This study will employ a descriptive cross-sectional study design, recruiting a clinic based MSM samples. This approach provides a clear picture of the population's characteristics and behaviors at a specific point, allowing for efficient data collection and prevalence assessment. The study aims to establish baseline data, explore associations, and generate hypotheses for future research or interventions within the MSM community.

Commented [AMM11]: Explain why this is the appropriate design for this study

Commented [WU12R11]: This study will employ a descriptive cross-sectional study design, recruiting a clinic based MSM samples. This approach provides a clear picture of the population's characteristics and behaviors at a specific point, allowing for efficient data collection and prevalence assessment. The study aims to establish baseline data, explore associations, and generate hypotheses for future research or interventions within the MSM community

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3.4 Sample size determination

Machakos County is a rapidly developing periurban area, reported to have a relatively high number of MSM seeking medical treatment in medical facilities (Kangethe et al., 2023). The county is home to 6 Dice Clinics which have been a crucial provider of medical care for MSM in recent years. The sample size will be calculated using Fisher *et al.*, 2021 and considering that the prevalence of HPV/ HIV coinfection among MSM in Kenya is 51.3%

$$N = Z^2 pq / d^2$$

Where N = Minimum sample size

Z = 1.96 standard error of 95% confidence

P = 0.513 (prevalence of the interest group this 51.3%)

d = 0.05 (inverse of 95% allowable error)

q = 1-p

This is calculated as follows

$$\frac{3.8416 \cdot 0.513 \cdot 10.487}{0.0025} = 384$$

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On average, 13 MSM are attended to daily at all the six (6) Dice Clinics in a month, and this study is expected to take three months. Therefore, the target population will be 1170 (13*30*3) MSM. Given that the target population is less than ten thousand, the size of sample (n) will be adjusted by applying the finite population correction factor in equation for the proportions:

$$n = \frac{no}{1 + \frac{(no - 1)}{N}}$$

$$n = \frac{384}{1 + \frac{(384 - 1)}{170}} = 118$$

Therefore, the desired minimum sample size will be **708**, 118 participants from each Dice clinic.

3.5 Sampling technique

The sampling technique that will be used in the selection of participants for this study will be random sampling. The sampling interval will be calculated based on a target a target population (sampling frame) of 1170 MSM in the three-month study period. Therefore, the study participant interval in each of the six Dice clinics will be 10 (1170/118), with the first participant picked randomly and the subsequent ones after every 10th MSM until the desired size is achieved.

Commented [AMM15]: Clarify this. Is it 13 msm per Dice daily?

Commented [WU16R15]: I have clarified that it is all Dice clinics

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3.6 Data and clinical sample collection

Data collection will employ a structured questionnaires issued to a client after signing the study consent form, to capture their socio-demographics and risk-factors-related data (Appendix IV and V). The study participant will be given instructions on HIV and HPV testing, and the result of the test indicated on the questionnaire. The samples to be collected will include anogenital swabs for HPV screening and blood samples corrected using rapid test kits for HIV screening. The swabs will be collected by the individual participants while the blood samples will be collected by medical staff at the testing center at the Dice Clinics. According to the MOH guidelines, HIV testing should be voluntary and conducted ethically in an environment where consent, confidentiality, counselling, correct results, connection (linkage) and creating an enabling environment can be assured (Joint Committee on Vaccination and Immunisation, 2015). The HPV swab and HIV blood samples will be stored in labeled containers, refrigerated or frozen, and transport with ice packs.

Commented [AMM17]: What kind of samples will be collected? Who will collect them? Will HIV counselors be engaged. What is MOH guidelines for HIV testing? How will the collected samples reach lab

Commented [WU18R17]: The samples to be collected will include anogenital swabs for HPV screening and blood samples corrected using rapid test kits for HIV screening. The swabs will be collected by the individual participants while the blood samples will be collected by medical staff at the testing center at the Dice Clinics. According to the MOH guidelines, HIV testing should be voluntary and conducted ethically in an environment where consent, confidentiality, counselling, correct results, connection (linkage) and creating an enabling environment can be assured (Joint Committee on Vaccination and Immunisation, 2015). The HPV swab and HIV blood samples will be stored in labeled containers, refrigerated or frozen, and transport with ice packs.

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3.7 Laboratory analysis

3.7.1 HPV molecular detection

The collected swabs will be are immersed in a lysis buffer containing Proteinase K (10 mg/ml), SDS (1%), Tris-HCl (100 mM, pH 8.0), and EDTA (50 mM). This lysis buffer facilitates cell membrane breakdown and the release of cellular components. Following a brief incubation period of approximately 30 minutes, the samples undergo centrifugation to separate proteins, leaving purified DNA ready for further analysis. Rigorous quality

control measures, including spectrophotometry and gel electrophoresis, are implemented to validate the accuracy and integrity of the extracted DNA.

DNA isolation will include the addition of a lysis buffer containing Proteinase K, SDS, Tris-HCl, and EDTA, followed by cell lysis, protein removal, and DNA purification. Measurements include Proteinase K (10 mg/ml), SDS (1%), Tris-HCl (100 mM, pH 8.0), EDTA (50 mM), guanidine hydrochloride (4 M), isopropanol (100%), and ethanol (70%). This extraction process takes approximately 60-90 minutes, ensuring the efficient recovery of high-quality genomic material for subsequent molecular analyses.

After DNA isolation, the identification of specific HPV genotypes will be achieved through genotyping assays. Targeted PCR amplification will involve the use of Tris-HCl (10 mM), EDTA (1 mM), guanidine hydrochloride, isopropanol, and ethanol. The precise measurements and steps in this stage will be critical, ensuring accurate identification. Genotyping by sequencing will follow, allowing for the comparison of sequences or hybridization patterns against established HPV genotype databases. Stringent controls are incorporated, and the entire genotyping process is typically completed within 24-48 hours, providing comprehensive insights into the viral landscape within the participant cohort.

3.7.2 HIV seroprevalence

In this study, HIV seroprevalence will be assessed using the standard HIV testing method conducted at Voluntary Counseling and Testing (VCT) centers. Rapid test kits will be used

Commented [AMM19]: Be specific on the test e.g. Determine and how positive cases will be confirmed based on MOH algorithm. Will you rely on professional counsellors. You should also specify that the HIV test will be done for the those with negative or unknown status as per clinic record.

Commented [WU20R19]: During the waiting period before the results are ready, counsellors will be available to offer counselling services to prepare the participant for the result.

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to detect HIV antibodies, and offer test results in minutes, with participants having been notified on how to read the results. The HIV test will be conducted on participants who are negative or whose status is unknown as per clinic records. Professional counsellors will be available to offering counselling to participants. Participants will provide blood samples, and the VCT testing protocol will employ rapid antibody tests, typically based on lateral flow immunochromatography. These tests commonly include components such as a sample pad, conjugate pad, reaction membrane, and absorbent pad. The procedure will involve applying a small blood sample to the test device, and if HIV antibodies are present, a visible line will appear on the test strip. The test results are usually available within 15-20 minutes. Quality control measures will be strictly followed, including the use of positive and negative controls, to ensure the reliability of the results. This method, widely utilized in VCT settings, allows for a quick and efficient assessment of HIV seroprevalence within the study population. The simplicity and rapid turnaround time make it a practical choice for large-scale seroprevalence studies. The timeframe for each VCT test, including sample collection and result interpretation, is expected to be within 30 minutes per participant. During the waiting period before the results are ready, counsellors will be available to offer counselling services to prepare the participant for the result.

???

3.7.3 HPV prevalence

Anogenital swabs will be collected from participants, who will do so in private and present samples ready for storage and screening. To ensure proper technique during collection, participants will be issued with vials and sterile swabs. A detailed guideline on how to safely extract a useful swab will be provided to the participants. The presented sample will

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- Commented [WU22R21]: Added as 3.7.3
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be stored at the clinic and transport to the Machakos Level 5 Hospital laboratory made every three days, to ensure correct storage conditions. Proper documentation and labeling will be adhered to.

3.8 Data management

In this study, the data analysis procedure will involve thorough data cleaning, processing, and analysis. Following the completion of factor analyses for both dependent and independent variable items, reliability analyses will be computed. Descriptive variables comprising percentage, range, mean, median and standards deviation will be determined for all. Proportions values shall also be compared with the use of Chi-square tests. A P-value will be considered statistically significant if its value is less than 0.05. Information on participants' code number, age, HPV and HIV status, marital status, circumcision status, employment status, level of education and number of life time sexual partners will be entered in MS Excel and organized in tables, bar graphs and charts. Correlation coefficients among independent and dependent variables will be calculated to explore the presence and characteristics of relationships between the study variables. The processing and analysis of data will be conducted using SPSS statistical software, ensuring a comprehensive examination of the study's variables and their interrelationships.

3.9 Ethical consideration

Ethical approval will be sought from Kenyatta University Ethical review committee, permit will be obtained from National Commission for Science, Technology and Innovation

(NACOSTI), and a permit from the Dice Clinics management. Informed consent will be given by the participants.

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Appendices

Appendix 1: Research work plan

Activity	Jan 2024	Feb 2024	Mar 2024	Apr 2024	May 2024	June 2024	July 2024
Proposal writing, presentation and submission							
Ethical approval							
Laboratory work, data collection and analysis							
Manuscript writing and publication							
Thesis writing and submission							
Defense							
Graduation							

Item	Quantity	Unit cost	Total
HPV cartilage	800	3000	240,000
HIV Kit	800	120	96,000
Swabs	800	10	8,000
Needles	800	10	8,000
Cotton wool	8(400g)	300	2,400
Methylated spirit	8(500ml)	250	2,000
Transport	90days	1000	90,000
Publication	2	70000	140,000
Thesis printing and binding	70	800	70,000
Personal			45,000+8000
Miscellaneous			50,000
Total			751,400

[illegible]

Appendix III: Study questionnaire

**Study title: HUMAN PAPILLOMA VIRUS AND HUMAN IMMUNODEFICIENCY
VIRUS INFECTIONS AMONG MEN WHO HAVE SEX WITH MEN IN
MACHAKOS COUNTY, KENYA**

Section 1: Participant demographics

Age: ☐ 18-24 ☐ 25-34 ☐ 35-44 ☐ 45-54 ☐ 55+

Sexual orientation

☐ Gay ☐ Bisexual
☐ Pansexual ☐ Queer
☐ Other (please specify): _____

Dice clinic you attend

☐ Mavoko ☐ Mwala
☐ Machakos ☐ Kalama
☐ Athiriver ☐ Tala
☐ Other (please specify): _____

Educational background

☐ High School or equivalent ☐ Some College/Technical School
☐ Bachelor's Degree ☐ Master's Degree
☐ Doctorate/Professional Degree

Section 2: Sexual health and behavior

Number of sexual partners in the last 6 months

☐ 0-1 ☐ 2-5
☐ 6-10 ☐ 11-20
☐ More than 20

Commented [AMM25]: correct the title and enrich this questionnaire

Commented [WU26R25]: done

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Consistent condom use

- ☐ Always ☐ Often
☐ Sometimes ☐ Rarely
☐ Never

History of sexually transmitted infections (STIs):

- ☐ Yes ☐ No ☐ Not Sure

Frequency of STI testing:

- ☐ Regularly ☐ Occasionally
☐ Rarely ☐ Never

Section 3: Knowledge and awareness

Awareness of HPV

- ☐ Yes ☐ No ☐ Not Sure

Awareness of HIV

- ☐ Yes ☐ No ☐ Not Sure

Sources of information (Select all that apply):

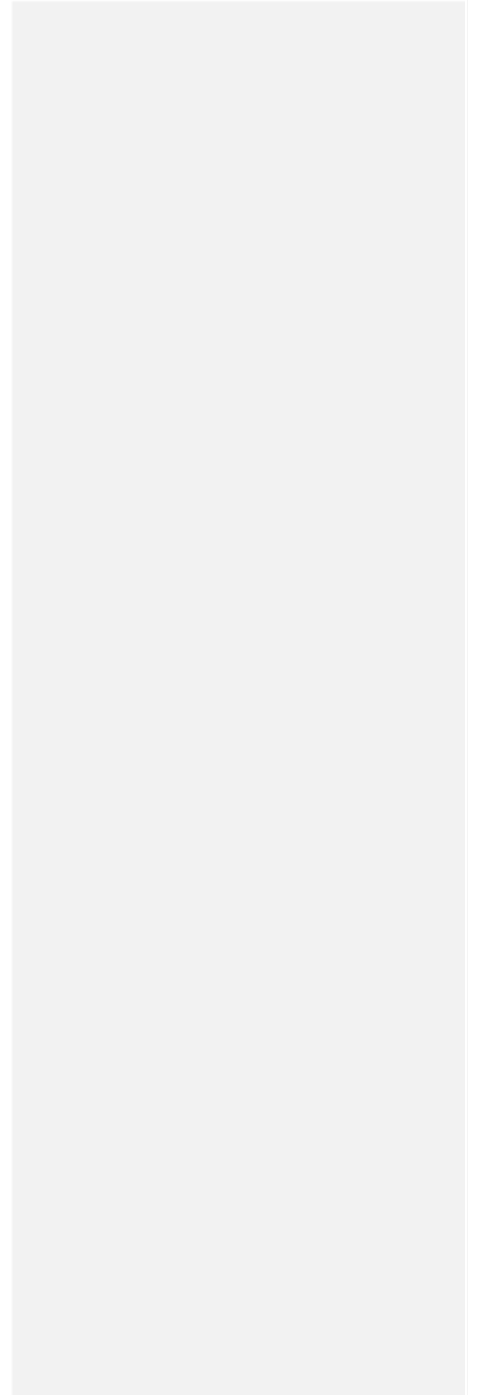
- ☐ Healthcare providers ☐ Educational campaigns
☐ Internet/Online resources ☐ Support groups
☐ Friends/Peers

Section 4: Medical history

HIV testing experience

- ☐ Never tested ☐ Tested and aware of status ☐ Tested but unaware of status

|



HPV Vaccination history

☐ Vaccinated ☐ Not vaccinated ☐ Not sure

Experience with HPV or HIV diagnosis

☐ Personal experience ☐ Partner's experience
☐ Friend's experience ☐ No direct experience

Section 5: Knowledge and access to healthcare

Access to LGBTQ+ friendly healthcare

☐ Yes ☐ No ☐ Not Sure

Barriers to seeking healthcare

☐ Stigma ☐ Lack of awareness
☐ Discrimination ☐ Cost
☐ Accessibility

Suggestions for improving healthcare access

Appendix IV: Information Sheet and Consent Form

I am Elizabeth Munee, a student at Kenyatta University undertaking a Masters course.

Title of my study is “HUMAN PAPILLOMA VIRUS AND HUMAN IMMUNODEFICIENCY VIRUS INFECTIONS AMONG MEN WHO HAVE SEX WITH MEN IN MACHAKOS COUNTY, KENYA”

Purpose of the study

This study is for academic purposes being conducted for research from Kenyatta University. It aims at identifying the epidemiology of HPV/HIV coinfection among men who have sex with men (MSM). The findings will guide in treatment and management of MSM and will inform on updating of policies relating to medical care for MSM. We are requesting for your assistance in this research, by responding to a few questions. We shall ask you to produce a HPV swab and HIV test for this study.

What will it involve for my participation?

If you consent to take part in this study, we shall need answers to a few questions which will help us to determine the risk factors linked to HPV/HIV coinfection. We shall request to you to provide us with a HPV swab and HIV test. You will not be required to take any additional tests apart from those related to ongoing treatment for the infection.

Are there any risks or advantages to me if I take part in the research?

There will be no risks to the patients because only a HPV swab and HIV test will be collected and it is a non-invasive procedure. Advantages are that the patient will be treated according to the individual HPV/HIV coinfection susceptibility results. It will guide on the need to upgrade prescription.

What will happen if I refuse to take part?

Taking part in research is ones wish and you have the free- will to choose whether to participate or not. If you agree now to participate or retreat at any time of the research, this will not interfere with your healthcare services now or in the future.

Who will be able to access my information in this research?

It is only the investigator and team involved in the research that will have access and view information of participants. To maintain participants' confidentiality, our research records will be stored safely in lockable drawers. Computers will be protected using password with limited sharing. Only patients' and laboratory numbers will be used

Benefits of the study

The information generated from this study will benefit patients because they will be treated according to the individual HPV/HIV coinfection susceptibility results. Patients' contacts will be used to inform them of the results in case they need any medical care interventions from the results. The clinicians will also benefit because the HPV/HIV coinfection trends will inform them of treatment options. The results will be released to the clinicians and then recorded in the patient's file. This information will also guide on the need to introduce HPV tests as part of the routine HIV test in MSM so that they use the susceptibility results

instead of empiric prescription in patient management. It will also improve on HPV surveillance to understand and minimize coinfection between HPV and HIV

What if I have any questions?

You can channel your questions at any time to any of our staff.

Elizabeth Munee - +254 725 420395

Dr. Abednego Musyoki - 0722588677

Consent Form

Study Title: **HUMAN PAPILLOMA VIRUS AND HUMAN IMMUNODEFICIENCY
VISUS INFECTIONS AMONG MEN WHO HAVE SEX WITH MEN IN
MACHAKOS COUNTY, KENYA**

I, being a participant _____ (participant's name) the
research has been explained to me and have understood all that have been read. My
questions have been answered satisfactorily. I understand that I can change my mind at any
stage and this will not affect the benefits due to me:

I hereby agree to take part in this research voluntarily.

Participant's signature: _____ Date

Participant's name: _____ Time

(Please print name)

I hereby confirm that I have followed all the study specific requirements and procedures in the SOP for obtaining informed consent.

Designee/investigator's signature: _____ **Date**

Designee/investigator's name: _____ **Time**

Only necessary if the Participant cannot read:

I attest that the information concerning this research has been accurately explained and clearly understood by the participant and that informed consent was freely given by the participant.

Witness' signature: _____ **Date**

Witness' name: _____ **Time**

*A witness is a person who is independent from the trial or a member of staff who was not involved in gaining the consent.

Thumbprint of the participant if they cannot write.

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