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ЕЖЕМЕСЯЧНЫЙ НАУЧНЫЙ ЖУРНАЛ

Медицинские новости Грузии

საქართველოს სამედიცინო სიახლენი

GEORGIAN MEDICAL NEWS

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GMN: Georgian Medical News is peer-reviewed, published monthly journal committed to promoting the science and art of medicine and the betterment of public health, published by the GMN Editorial Board since 1994. GMN carries original scientific articles on medicine, biology and pharmacy, which are of experimental, theoretical and practical character; publishes original research, reviews, commentaries, editorials, essays, medical news, and correspondence in English and Russian.

GMN is indexed in MEDLINE, SCOPUS, PubMed and VINITI Russian Academy of Sciences. The full text content is available through EBSCO databases.

GMN: Медицинские новости Грузии - ежемесячный рецензируемый научный журнал, издаётся Редакционной коллегией с 1994 года на русском и английском языках в целях поддержки медицинской науки и улучшения здравоохранения. В журнале публикуются оригинальные научные статьи в области медицины, биологии и фармации, статьи обзорного характера, научные сообщения, новости медицины и здравоохранения. Журнал индексируется в MEDLINE, отражён в базе данных SCOPUS, PubMed и ВИНИТИ РАН. Полнотекстовые статьи журнала доступны через БД EBSCO.

GMN: Georgian Medical News – საქართველოს სამედიცინო სიახლენი – არის ყოველთვიური სამეცნიერო სამედიცინო რეცენზირებადი ჟურნალი, გამოიცემა 1994 წლიდან, წარმოადგენს სარედაქციო კოლეგიისა და აშშ-ის მეცნიერების, განათლების, ინდუსტრიის, ხელოვნებისა და ბუნებისმეტყველების საერთაშორისო აკადემიის ერთობლივ გამოცემას. GMN-ში რუსულ და ინგლისურ ენებზე ქვეყნდება ექსპერიმენტული, თეორიული და პრაქტიკული ხასიათის ორიგინალური სამეცნიერო სტატიები მედიცინის, ბიოლოგიისა და ფარმაციის სფეროში, მიმოხილვითი ხასიათის სტატიები.

ჟურნალი ინდექსირებულია MEDLINE-ის საერთაშორისო სისტემაში, ასახულია SCOPUS-ის, PubMed-ის და ВИНИТИ РАН-ის მონაცემთა ბაზებში. სტატიების სრული ტექსტი ხელმისაწვდომია EBSCO-ს მონაცემთა ბაზებიდან.

WEBSITE

www.geomednews.com

К СВЕДЕНИЮ АВТОРОВ!

При направлении статьи в редакцию необходимо соблюдать следующие правила:

1. Статья должна быть представлена в двух экземплярах, на русском или английском языках, напечатанная через **полтора интервала на одной стороне стандартного листа с шириной левого поля в три сантиметра**. Используемый компьютерный шрифт для текста на русском и английском языках - **Times New Roman** (**Кириллица**), для текста на грузинском языке следует использовать **AcadNusx.** Размер шрифта - **12**. К рукописи, напечатанной на компьютере, должен быть приложен CD со статьей.

2. Размер статьи должен быть не менее десяти и не более двадцати страниц машинописи, включая указатель литературы и резюме на английском, русском и грузинском языках.

3. В статье должны быть освещены актуальность данного материала, методы и результаты исследования и их обсуждение.

При представлении в печать научных экспериментальных работ авторы должны указывать вид и количество экспериментальных животных, применявшиеся методы обезболивания и усыпления (в ходе острых опытов).

4. К статье должны быть приложены краткое (на полстраницы) резюме на английском, русском и грузинском языках (включающее следующие разделы: цель исследования, материал и методы, результаты и заключение) и список ключевых слов (key words).

5. Таблицы необходимо представлять в печатной форме. Фотокопии не принимаются. **Все цифровые, итоговые и процентные данные в таблицах должны соответствовать таковым в тексте статьи**. Таблицы и графики должны быть озаглавлены.

6. Фотографии должны быть контрастными, фотокопии с рентгенограмм - в позитивном изображении. Рисунки, чертежи и диаграммы следует озаглавить, пронумеровать и вставить в соответствующее место текста **в tiff формате**.

В подписях к микрофотографиям следует указывать степень увеличения через окуляр или объектив и метод окраски или импрегнации срезов.

7. Фамилии отечественных авторов приводятся в оригинальной транскрипции.

8. При оформлении и направлении статей в журнал МНГ просим авторов соблюдать правила, изложенные в «Единых требованиях к рукописям, представляемым в биомедицинские журналы», принятых Международным комитетом редакторов медицинских журналов - http://www.spinesurgery.ru/files/publish.pdf и http://www.nlm.nih.gov/bsd/uniform_requirements.html
В конце каждой оригинальной статьи приводится библиографический список. В список литературы включаются все материалы, на которые имеются ссылки в тексте. Список составляется в алфавитном порядке и нумеруется. Литературный источник приводится на языке оригинала. В списке литературы сначала приводятся работы, написанные знаками грузинского алфавита, затем кириллицей и латиницей. Ссылки на цитируемые работы в тексте статьи даются в квадратных скобках в виде номера, соответствующего номеру данной работы в списке литературы. Большинство цитированных источников должны быть за последние 5-7 лет.

9. Для получения права на публикацию статья должна иметь от руководителя работы или учреждения визу и сопроводительное отношение, написанные или напечатанные на бланке и заверенные подписью и печатью.

10. В конце статьи должны быть подписи всех авторов, полностью приведены их фамилии, имена и отчества, указаны служебный и домашний номера телефонов и адреса или иные координаты. Количество авторов (соавторов) не должно превышать пяти человек.

11. Редакция оставляет за собой право сокращать и исправлять статьи. Корректура авторам не высылается, вся работа и сверка проводится по авторскому оригиналу.

12. Недопустимо направление в редакцию работ, представленных к печати в иных издательствах или опубликованных в других изданиях.

При нарушении указанных правил статьи не рассматриваются.

REQUIREMENTS

Please note, materials submitted to the Editorial Office Staff are supposed to meet the following requirements:

1. Articles must be provided with a double copy, in English or Russian languages and typed or compu-ter-printed on a single side of standard typing paper, with the left margin of **3** centimeters width, and **1.5** spacing between the lines, typeface - **Times New Roman** (**Cyrillic**), print size - **12** (referring to Georgian and Russian materials). With computer-printed texts please enclose a CD carrying the same file titled with Latin symbols.

2. Size of the article, including index and resume in English, Russian and Georgian languages must be at least 10 pages and not exceed the limit of 20 pages of typed or computer-printed text.

3. Submitted material must include a coverage of a topical subject, research methods, results, and review.

Authors of the scientific-research works must indicate the number of experimental biological species drawn in, list the employed methods of anesthetization and soporific means used during acute tests.

4. Articles must have a short (half page) abstract in English, Russian and Georgian (including the following sections: aim of study, material and methods, results and conclusions) and a list of key words.

5. Tables must be presented in an original typed or computer-printed form, instead of a photocopied version. **Numbers, totals, percentile data on the tables must coincide with those in the texts of the articles.** Tables and graphs must be headed.

6. Photographs are required to be contrasted and must be submitted with doubles. Please number each photograph with a pencil on its back, indicate author's name, title of the article (short version), and mark out its top and bottom parts. Drawings must be accurate, drafts and diagrams drawn in Indian ink (or black ink). Photocopies of the X-ray photographs must be presented in a positive image in **tiff format**.

Accurately numbered subtitles for each illustration must be listed on a separate sheet of paper. In the subtitles for the microphotographs please indicate the ocular and objective lens magnification power, method of coloring or impregnation of the microscopic sections (preparations).

7. Please indicate last names, first and middle initials of the native authors, present names and initials of the foreign authors in the transcription of the original language, enclose in parenthesis corresponding number under which the author is listed in the reference materials.

8. Please follow guidance offered to authors by The International Committee of Medical Journal Editors guidance in its Uniform Requirements for Manuscripts Submitted to Biomedical Journals publication available online at: http://www.nlm.nih.gov/bsd/uniform_requirements.html
http://www.icmje.org/urm_full.pdf
In GMN style for each work cited in the text, a bibliographic reference is given, and this is located at the end of the article under the title "References". All references cited in the text must be listed. The list of references should be arranged alphabetically and then numbered. References are numbered in the text [numbers in square brackets] and in the reference list and numbers are repeated throughout the text as needed. The bibliographic description is given in the language of publication (citations in Georgian script are followed by Cyrillic and Latin).

9. To obtain the rights of publication articles must be accompanied by a visa from the project instructor or the establishment, where the work has been performed, and a reference letter, both written or typed on a special signed form, certified by a stamp or a seal.

10. Articles must be signed by all of the authors at the end, and they must be provided with a list of full names, office and home phone numbers and addresses or other non-office locations where the authors could be reached. The number of the authors (co-authors) must not exceed the limit of 5 people.

11. Editorial Staff reserves the rights to cut down in size and correct the articles. Proof-sheets are not sent out to the authors. The entire editorial and collation work is performed according to the author's original text.

12. Sending in the works that have already been assigned to the press by other Editorial Staffs or have been printed by other publishers is not permissible.

Articles that Fail to Meet the Aforementioned Requirements are not Assigned to be Reviewed.

ავტორთა საყურადღებოდ!

რედაქციაში სტატიის წარმოდგენისას საჭიროა დავიცვათ შემდეგი წესები:

1. სტატია უნდა წარმოადგინოთ 2 ცალად, რუსულ ან ინგლისურ ენებზე,დაბეჭდილი სტანდარტული ფურცლის 1 გვერდზე, 3 სმ სიგანის მარცხენა ველისა და სტრიქონებს შორის 1,5 ინტერვალის დაცვით. გამოყენებული კომპიუტერული შრიფტი რუსულ და ინგლისურენოვან ტექსტებში - **Times New Roman (Кириллица)**, ხოლო ქართულენოვან ტექსტში საჭიროა გამოვიყენოთ **AcadNusx.** შრიფტის ზომა – 12. სტატიას თან უნდა ახლდეს CD სტატიით.

2. სტატიის მოცულობა არ უნდა შეადგენდეს 10 გვერდზე ნაკლებს და 20 გვერდზე მეტს ლიტერატურის სიის და რეზიუმეების (ინგლისურ, რუსულ და ქართულ ენებზე) ჩათვლით.

3. სტატიაში საჭიროა გაშუქდეს: საკითხის აქტუალობა; კვლევის მიზანი; საკვლევი მასალა და გამოყენებული მეთოდები; მიღებული შედეგები და მათი განსჯა. ექსპერიმენტული ხასიათის სტატიების წარმოდგენისას ავტორებმა უნდა მიუთითონ საექსპერიმენტო ცხოველების სახეობა და რაოდენობა; გაუტკივარებისა და დაძინების მეთოდები (მწვავე ცდების პირობებში).

4. სტატიას თან უნდა ახლდეს რეზიუმე ინგლისურ, რუსულ და ქართულ ენებზე არანაკლებ ნახევარი გვერდის მოცულობისა (სათაურის, ავტორების, დაწესებულების მითითებით და უნდა შეიცავდეს შემდეგ განყოფილებებს: მიზანი, მასალა და მეთოდები, შედეგები და დასკვნები; ტექსტუალური ნაწილი არ უნდა იყოს 15 სტრიქონზე ნაკლები) და საკვანძო სიტყვების ჩამონათვალი (key words).

5. ცხრილები საჭიროა წარმოადგინოთ ნაბეჭდი სახით. ყველა ციფრული, შემაჯამებელი და პროცენტული მონაცემები უნდა შეესაბამებოდეს ტექსტში მოყვანილს.

6. ფოტოსურათები უნდა იყოს კონტრასტული; სურათები, ნახაზები, დიაგრამები - დასათაურებული, დანომრილი და სათანადო ადგილას ჩასმული. რენტგენოგრამების ფოტოასლები წარმოადგინეთ პოზიტიური გამოსახულებით **tiff** ფორმატში. მიკროფოტოსურათების წარწერებში საჭიროა მიუთითოთ ოკულარის ან ობიექტივის საშუალებით გადიდების ხარისხი, ანათალების შეღებვის ან იმპრეგნაციის მეთოდი და აღნიშნოთ სურათის ზედა და ქვედა ნაწილები.

7. სამამულო ავტორების გვარები სტატიაში აღინიშნება ინიციალების თანდართვით, უცხოურისა – უცხოური ტრანსკრიპციით.

8. სტატიას თან უნდა ახლდეს ავტორის მიერ გამოყენებული სამამულო და უცხოური შრომების ბიბლიოგრაფიული სია (ბოლო 5-8 წლის სიღრმით). ანბანური წყობით წარმოდგენილ ბიბლიოგრაფიულ სიაში მიუთითეთ ჯერ სამამულო, შემდეგ უცხოელი ავტორები (გვარი, ინიციალები, სტატიის სათაური, ჟურნალის დასახელება, გამოცემის ადგილი, წელი, ჟურნალის №, პირველი და ბოლო გვერდები). მონოგრაფიის შემთხვევაში მიუთითეთ გამოცემის წელი, ადგილი და გვერდების საერთო რაოდენობა. ტექსტში კვადრატულ ფჩხილებში უნდა მიუთითოთ ავტორის შესაბამისი N ლიტერატურის სიის მიხედვით. მიზანშეწონილია, რომ ციტირებული წყაროების უმეტესი ნაწილი იყოს 5-6 წლის სიღრმის.

9. სტატიას თან უნდა ახლდეს: ა) დაწესებულების ან სამეცნიერო ხელმძღვანელის წარდგინება, დამოწმებული ხელმოწერითა და ბეჭდით; ბ) დარგის სპეციალისტის დამოწმებული რეცენზია, რომელშიც მითითებული იქნება საკითხის აქტუალობა, მასალის საკმაობა, მეთოდის სანდოობა, შედეგების სამეცნიერო-პრაქტიკული მნიშვნელობა.

10. სტატიის ბოლოს საჭიროა ყველა ავტორის ხელმოწერა, რომელთა რაოდენობა არ უნდა აღემატებოდეს 5-ს.

11. რედაქცია იტოვებს უფლებას შეასწოროს სტატია. ტექსტზე მუშაობა და შეჯერება ხდება საავტორო ორიგინალის მიხედვით.

12. დაუშვებელია რედაქციაში ისეთი სტატიის წარდგენა, რომელიც დასაბეჭდად წარდგენილი იყო სხვა რედაქციაში ან გამოქვეყნებული იყო სხვა გამოცემებში.

აღნიშნული წესების დარღვევის შემთხვევაში სტატიები არ განიხილება.

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MOLECULAR DETECTION OF HIGH RISK HUMAN PAPILLOMA VIRUS SUBTYPES IN CERVICAL SMEARS AMONG SUDANESE WOMEN

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Abstract.

Background: Cervical cancer is the fourth most frequent cancer in women worldwide, with an expected 604,000 new cases and 342,000 deaths in 2020. The human papillomavirus is recognized as a primary cause of cervical cancer. This study aims to elucidate the distribution of human papillomavirus (HPV) subtypes and examine their correlation with cytological abnormalities among Sudanese women, providing critical insights for targeted vaccination strategies and cervical cancer prevention programs.

Materials and Method: This descriptive cross-sectional laboratory-based investigation was carried out in Soba Teaching Hospital and Academy Teaching Hospital in Khartoum, Sudan. This study included 236 Sudanese female patients who visited the clinic for follow-up or screening. The Pap smear and HPV DNA screening tests for 16,18,33,45, 58 were conducted. The data was analyzed using the Statistical Package for Social Sciences, version 26.0.

Results: Out of 236 cervical smear samples from Sudanese females ranging in age from 20 to 80, with a mean of 37.7 years, 99 (41.9%) were positive and 137 (58.1%) were negative. High-risk HPV subtypes included HPV16,18,33,45, 58, with percentages of 5.1%, 2.5%, 24.2%, 7.6%, and 2.5%. High-risk HPV subtypes were found in 99 of 135 abnormal pap smears, with a significant p-value of 0.001.

Conclusion: High-risk human papillomavirus subtypes play an important role in cervical cancer occurrence. High-risk HPV 33 was the predominant subtype detected. Further nationide study is required fous on the prevalence of HPV-genotype 33, and distribution of other high-risk genotypes in the country regions specially after the refugee return back home from the east african regional countries.

Key words. HPV, PCR, cervical smears, cervical cancer, Sudan.

Introduction.

Cervical cancer is a major public health issue worldwide, with an estimated 662,000 new cases and 348,000 deaths reported in 2022 [1-3]. It is the fourth most frequent cancer in women worldwide, with the highest burden seen in low- and middle-income countries, particularly Sub-Saharan Africa, South-East Asia, and Central America [1].

These locations have significant gaps in healthcare access, including poor access to HPV vaccination, cervical cancer screening, and treatment services [1]. Persistent infection with high-risk strains of human papillomavirus (HPV) is the leading cause of cervical cancer, which is exacerbated by HIV infection, early sexual engagement, and smoking [2].

In contrast, high-income nations report lower incidence and death as a result of widespread HPV vaccination and screening programs [1,2]. Addressing this inequality through preventative strategies, such as global access to vaccine and screening, is critical for lowering the cervical cancer burden, particularly in poor areas [1,3].

The prevalence of cervical cancer in Sudan highlights serious public health challenges, mainly due to inadequate access to preventive measures such as HPV vaccination and organised screening programs. Sudan has an estimated 13.8 million women aged 15 and up who are at risk of having cervical cancer. According to current data, around 1,227 new instances of cervical cancer are detected each year, with 828 women dying as a result. The age-standardized incidence rate is 8.7 per 100,000 women, while the mortality rate is 6.1 per 100,000 women [3].

Cervical cancer is the second most frequent cancer in Sudan, ranking third among women aged 15 to 44 [4,5]. Furthermore, HPV types 16 and 18, the high-risk strains most typically associated with cervical cancer, account for approximately 78.9% of all invasive cervical malignancies in the region [3].

While specific national data on HPV prevalence are unavailable, regional estimates indicate that approximately 3% of women in Northern Africa carry HPV 16/18 infections at any given time [4]. Sudan has an established HPV vaccine program and no national cervical cancer screening guidelines, which severely limits prevention efforts [3].

This emphasises the critical need for policy actions, including as vaccination and screening strategies, to lower the burden of

cervical cancer in Sudan.

This study investigates the prevalence and genotype distribution of human papillomavirus (HPV) among women with suspected cervical lesions in Sudan, while evaluating associations between specific subtypes and cytopathological findings. The findings will inform development of evidence-based cervical cancer screening guidelines and HPV vaccination policies, addressing a critical gap in Sudan's current preventive healthcare framework where structured immunization programs remain absent despite the disease's high burden.

Materials and Methods.

After receiving approval from the Institutional Ethical Committee, a cross-sectional study was carried out on 236 women who met the inclusion criteria and were enrolled in the Omdurman Cervical Cancer Prevention Centre, and the expected sample size was 256 based on the HPV information center, 2023 [3].

Consecutive, sexually active women aged 18 and older with various complaints of vaginal discharge, bleeding, itching, or without symptoms who agreed to undergo speculum examination and planned investigations were included in the study. Pregnancy, women who had recently (within the last 15 days) received antibiotics or used intravaginal medicines, and menstruation were all excluded.

A written and informed permission was obtained, and each participant was asked to complete a brief questionnaire about their demographic information, including socioeconomic position, urogenital infections, medical history, sexual activities, and smoking, followed by a clinical evaluation using the predesigned case record form. Cervical samples were taken for traditional Pap smears.

The Ayre's spatula and cytobrush used for cervical cytology were immediately transferred to 15-ml plastic vials containing 5 ml of sterile phosphate-buffered saline and maintained at -70°C in a deep freezer until isolation of DNA for polymerase chain reaction (PCR) assay for HPV.

A pathologist with experience reading cervical cytology specimens, who was blinded to clinical details, participated in the trial and reviewed the cytosmears. Cervical smears were diagnosed using the Bethesda classification. PCR was used to detect HPV.

HPV-DNA Extraction and Genotyping:

Viral HPV-DNA was extracted from cervical scrapes using a commercial kit (Qiagen, Germany) according to the manufacturer's protocol. For genotyping, PCR was carried out in a 25 µL reaction mixture containing 5 µL of DNA, 2 µL each of forward and reverse primers (10 pmol/µL), 5 µL of 2 mM dNTPs, 25 mM $MgCl_2$, 2.5 U Taq polymerase, 1× PCR buffer (Intron Biotechnology, Korea), and 13 µL nuclease-free ddH_2O. The thermal cycling protocol consisted of an initial denaturation at 94°C for 5 min, followed by 40 cycles of denaturation (94°C, 1 min), annealing (60°C, 1 min), and extension (72°C, 2 min), with a final extension at 72°C for 5 min. Genotyping was performed using type-specific primers targeting HPV-16, 18, 58, 45, and 33 (table 1). The resulting amplicons were separated by agarose gel electrophoresis, visualized under UV light (Uni EQUIP, Germany), and compared against a 100 bp DNA ladder for size-based genotype determination.

The assay employed two optimized primer mixtures for concurrent amplification: Set I targeting HPV-45 and HPV-16, and Set II detecting HPV-18, HPV-33, and HPV-58, with β-actin incorporated as an endogenous control in both reaction sets. Primer concentrations were carefully balanced to minimize competitive inhibition, while uniform annealing temperatures were maintained across all targets to ensure amplification efficiency. This dual-set design enabled comprehensive genotyping while controlling for amplification integrity through the constitutive β-actin reference.

Results.

This descriptive cross-sectional laboratory-based investigation was carried out at Soba Teaching Hospital and the Academy Teaching Hospital in Khartoum, Sudan.

This study included 236 Sudanese female patients who visited the clinic for follow-up or screening. A questionnaire was devised to collect demographic and clinical information, as well as other study-related data. Descriptive statistics were employed to characterize the distribution of HPV genotypes, cytological classifications (Bethesda system), and clinical complaint frequencies. Categorical variables, including HPV genotypes, cytological findings, and clinical presentations, were compared using cross-tabulation with chi-square tests in (IBM SPSS Statistics 21) to assess associations between these parameters across participant subgroups.

The bulk of the participants (64.9%) are fertilized, sexually active women between the ages of 20 and 40, while 35.1% are menopausal women/stages with an average age of 37.7 (Table 2 and Figure 1).

The most common cytological diagnoses among individuals were NILM (42.8%), ASCUS (3.4%), LSIL (28.4%), and HSIL (20.3%), with adenocarcinoma and SCC accounting for just 2.5% each (Table 3).

In this investigation, the total burden of HR HPV infection was 99/236 (41.9%), with 137/236 (58.1%) being negative. Among the HR HPV-positive cases, 16 (57%) had the HR HPV 33 genotype, whereas 12 (43%) had other HR HPV genotypes (16, 18, 45, and 58) (Table 6).

Asymptomatic participants made up 43.2% (32), followed by bleeding at 27% (20), vaginal discharge at 18.9% (14), irregular cycle at 8.1% (5), and polyp at 0.07% (2).

Asymptomatic participants included 8 LSIL, 8 HSIL, and 2 ASCUS, with bleeding being the most common symptom for SCC and adenocarcinoma (Table 4).

There was no significant correlation between cytological findings and age groups and clinical complaints ($p = 0.18$). There was a significant correlation between HR HPV genotypes and age groups (p-value = 0.018). 16/28 (57%) were found in the 20-40 age group, and 12/28 (43%) in the 41-80 age group (Table 5).

This investigation discovered the most common HR HPV genotype, 33, which was found in 19 LSIL patients and 36 HSIL cases. Other HR HPV genotypes include HR HPV genotype 16 in 6 instances of SCC and HR HPV genotype 18 in 6 cases of Adenocarcinoma.

Table 1. *HPV genotype-specific primer sequences and corresponding amplicon characteristics.*

Primers	**Primer Name**	**Sequence (5'→3')**	**PCR Product (bp)**
I*	HPV-16 L1F	CACTATTTTGGAGGACTGGAAT	291
	HPV-16 L1R	GATGAGGTGGTGGGTGTAGC	
	HPV-45 L1F	TTTTATCATGCAGGCAGTTCC	233
	HPV-45 L1R	CCACGACCAATTTCCATACC	
II*	HPV-18 L1F	GCCCCTGCCTCTACACAGTA	292
	HPV-18 L1R	ATCCTGCTTATTGCCACCAC	
	HPV-33 L1F	TAACACCTCCTCCATCTGCT	202
	HPV-33 L1R	CCTGCCTGTAATAAAAACTTGC	
	HPV-58 L1F	GATTTGTTACCTCCCAGGCTATT	233
	HPV-58 L1R	CTTTTTGCGTTTGGTGGATG	
Control	β-actin F	AGCCATGTACGTTGCTATCC	498
	β-actin R	TTGGCGTACAGGTCTTTGC	

**Two sets of primers. Sets (I) and (II) were used. Each mixture also included the b-actin primer set as an internal control.*

Table 2. *Demographic distribution of study participants by age group.*

Age groups	Frequency	Percent	Mean
20-40	**154**	**65.3%**	37.7838
41-80	82	34.7%	
Total	236	100.0%	

Table 3. *Distribution of cervical cytology results classified by the Bethesda System.*

Diagnosis	Frequency	Percent
NILM	**101**	**42.8**
ASCUS	8	3.4
LSIL	**67**	**28.4**
HSIL	**48**	**20.3**
SCC	6	2.5
Adeno	6	2.5
Total	236	100.0

Chi-Square Tests= .000 significant p. value

Table 4. *Association between Bethesda cytology diagnosis and HPV detection frequency.*

Diagnosis	**HPV positive**	**HPV negative**	**Total**
NILM	0	101	101
ASCUS	2	6	8
LSIL	**37**	30	**67**
HSIL	**48**	0	**48**
SCC	6	0	6
Adeno	6	0	6
Total	99	137	236

Table 5. *Age-stratified distribution of high-risk HPV genotypes among study participants.*

HPV Genotypes	**Age groups**		**Total**
	20-40	41-80	
16	0	**12**	12
18	6	0	6
33	**43**	**14**	**57**
45	12	6	18
58	0	6	6
None	93	44	137
Total	154	82	236

Table 6. *Stratified association between Bethesda cytology diagnoses and high-risk HPV subtype distribution.*

HPV Subtypes	NILM	ASCUS	LSIL	HSIL	SCC	Adeno	Percent	Total
16	0	0	6	0	6	0	5.1	12
18	0	0	0	0	0	6	2.5	6
33	0	**2**	**19**	**36**	0	0	**24.2**	**57**
45	0	0	6	12	0	0	7.6	18
58	0	0	6	0	0	0	2.5	6
none	101	6	30	0	0	0	58.1	137
Total	101	8	67	48	6	6	100.0	236

Chi-Square Tests= 0.001 significant p. value

Table 7. *Association between high-risk HPV subtypes and presenting clinical complaints among study participants.*

HPV subtypes	Bleeding	Discharg	Irregular cycle	Polyps	No symptoms	Total
16	12	0	0	0	0	12
18	6	0	0	0	0	6
33	**12**	**12**	**6**	0	**27**	**57**
45	2	0	0	0	16	18
58	0	0	0	0	6	6
None	30	27	12	6	62	137
Total	62	39	18	6	111	236

p.value = 0.18

Table 8. *Association between presenting clinical complaints and Bethesda cytological diagnoses among study participants.*

Diagnosis							
Complaint	**NILM**	**ASCUS**	**LSIL**	**HSIL**	**SCC**	**adeno**	**Total**
Bleeding	30	0	8	12	6	6	62
Discharg	9	0	24	6	0	0	39
Irregular cycle	12	0	0	6	0	0	18
Polyps	6	0	0	0	0	0	6
No symptoms	44	8	35	24	0	0	111
Total	101	8	67	48	6	6	236

p.value = 0.018

HR HPV genotype 45 was found in 6 cases of HSIL, 12 in LSIL, and 58 in 6 cases of LSIL. HR HVP genotypes were not found in NILM. The cross-tabulation revealed a statistically significant correlation with a p-value of 0.001.

The study participants' ages ranged from 20 to 80 years, with the majority (65.3%) falling into the 20–40-year age group and 34.7% falling between 41 and 80 years. The mean age of the participants was 37.78 years.

According to the Bethesda system, 42.8% of participants had Negative for Intraepithelial Lesion or Malignancy (NILM), 3.4% had Atypical Squamous Cells of Undetermined Significance (ASCUS), 28.4% had Low-Grade Squamous Intraepithelial Lesions (LSIL), 20.3% had High-Grade Squamous Intraepithelial Lesions (HSIL), and 2.5% had either Squamous Cell Carcinoma (SCC) or adenocarcinoma (Adeno) (Figures 2 & 3).

The correlation between cervical cytological patterns and high-risk human papillomavirus (HR HPV) identification was significant. Among the participants, 41.9% tested positive for HPV, while 58.1% tested negative. HPV subtype 33 was the most commonly detected (24.2%), followed by subtype 45 (7.6%), subtype 16 (5.1%), and subtype 18 (2.5%).

A cross-tabulation study revealed that all NILM cases tested negative for HPV, but 37 of 67 LSIL cases and all 48 HSIL cases tested positive for HPV. All cases of SCC and adenocarcinoma were HPV-positive.

In terms of the relationship between Bethesda diagnoses and HPV subtypes, subtype 33 was most prevalent in LSIL and HSIL cases, whereas subtype 45 was detected in HSIL cases. Subtype 16 was found in both LSIL and adenocarcinoma cases, but subtype 18 was only seen in adenocarcinoma patients (Table 6).

Further examination of age distribution and HPV subtype revealed that participants aged 20 to 40 had a higher incidence of HPV subtype 33 than those aged 41 to 80. Participants' complaints varied, with the majority reporting bleeding (26.3%), discharge (16.5%), irregular periods (7.6%), and polyps (2.5%). Interestingly, 47% of individuals reported having no symptoms at all (Table 7).

There was also a strong link between complaints and Bethesda diagnosis. For example, those with HSIL frequently reported bleeding, but those with LSIL were more likely to present with discharge. The association was statistically significant, with a p-value of 0.018 (Table 8).

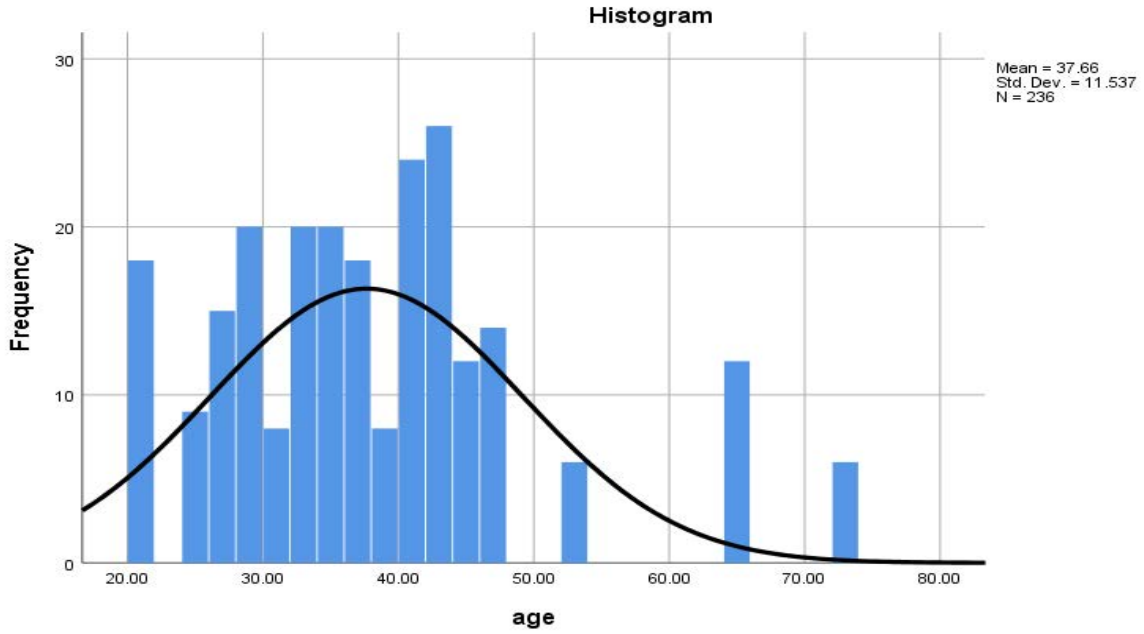

Figure 1. *Age distribution among study participants.*

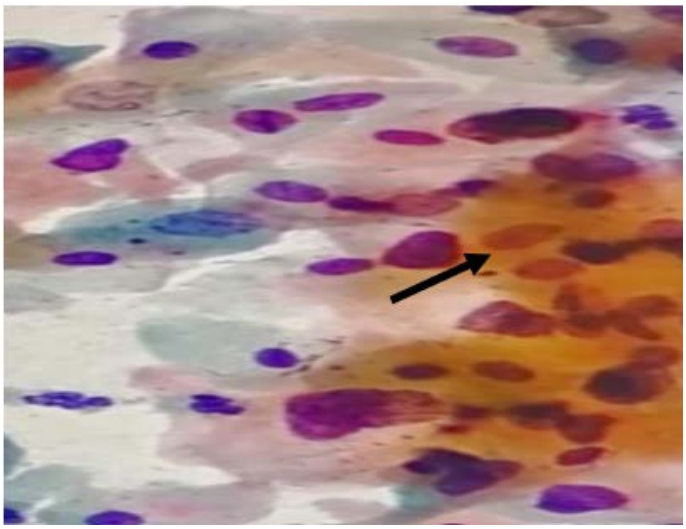

Figure 2. *Histopathological features of high-grade squamous intraepithelial lesion (HSIL) X 40 Pap stain.*

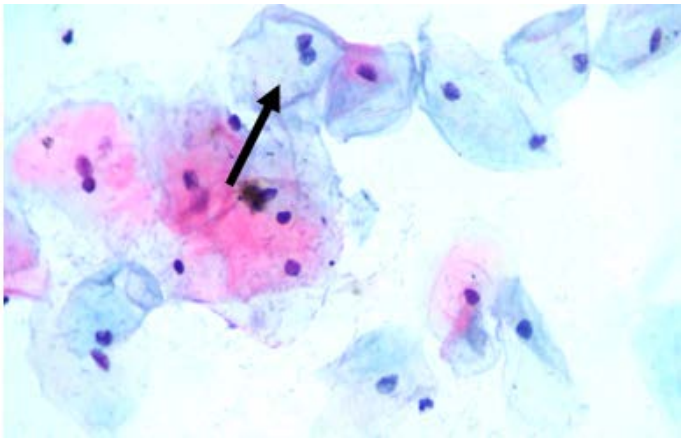

Figure 3. *Cytomorphologic features of low-grade squamous intraepithelial lesion (LSIL) X 40 Pap stain.*

Discussion.

This study examined the prevalence of HR HPV and cervical cytology abnormalities in two Obstetrics and Gynaecology and Reproductive Health clinics in Khartoum, Sudan.

The findings of this study offer a new perspective on the frequency and genotypes of high-risk human papillomavirus (HR HPV) among Sudanese women. The most common cytological diagnoses among participants were NILM at 45.9%, ASCUS at 2.7%, LSIL at 24.3%, and HSIL at 21.6%, with adenocarcinoma and SCC accounting for just 2.7% each (Table 2).

These findings are consistent with the study conducted in India, as well as the findings found in both studies by Mohan and Karthika [6,7]: 64.8% were negative for intraepithelial malignancy, and 17.2% showed benign inflammatory changes. They also agree to some extent with Ali et al, who conducted a study in Ethiopia and found that Pap smear abnormalities were observed in 13.1% (48/366) of study subjects [8]. Among the abnormalities, 3 (6.3%), 39 (81.3%), and 6 (12.5%) were ASCUS, LSIL, and HSIL, respectively.

In terms of the relationship between clinical symptoms and cytological findings, the majority of women were asymptomatic (43.2%), with 27% having bleeding and 18.9% having vaginal discharge.

Furthermore, 8 cases of LSIL, 8 cases of HSIL, and 2 cases of ASCUS arrived at the gynaecologic clinic with no symptoms, emphasising the significance of regular cervical cancer screening programs using Pap tests. These findings contrast with the findings of Wong LP et al. [9], who found that vaginal discharge was the most common complaint, affecting 36.96% of the participants.

The overall burden of HR HPV infection in this study was 28/74, (37.8%). Among the HR HPV-positive cases, 16 (57%) had the HR HPV 33 genotype, whereas 12 (43%) had other HR HPV (HR HPV genotypes 16 4 cases (5.4%), HR HPV 18 2 cases (2.7%), HR HPV 45 4 cases (5.4%), and HR HPV 58 4 cases (5.4%). The HR HPV was not found in 46 (62.2%) of the study participants.

The positive cases of HR HPV are higher than the study conducted in Ethiopia by Ali et al. [8], who found that the overall HR HPV burden was 13.7%. "Other HR HPV" genotypes (31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, or 68 types) were the most frequent (76%) genotypes identified in their study, followed by HR HPV 16 (16%). This difference might be because the participants in this study were women with cervical complaints, and all samples were cases of cervical dysplasia, which may result in higher values.

Our data exceeded the anticipated prevalence reported for all HPV genotypes in Sub-Saharan African nations (21.8%) [5] and Nigeria (21.6%) [8]. In other studies, conducted in Ethiopia [8,10] found that HR HPV 16 is the most common kind. In contrast, the most common genotype found in the current investigation was HR HPV" genotype 33, which contributed 57%, followed by HR HPV 16 (5.4%).

Our findings are consistent with a worldwide meta-analysis assessment [5], which found that the most common genotype in Eastern Africa was HR HPV 52, followed by HPV 16. Another study [10] discovered that HPV-positive women in sub-Saharan Africa were less likely to be infected with HPV 16 than women in Europe.

Similarly, another study [11] in South Korea found that (54.9%) infected with the most genotypes were HPV 52 (16.8%) and 58 (20.4%). Geographic variation and host immunogenic variables could explain the variance in genotype frequency seen across investigations.

An age-specific HPV infection research in South Africa [6] indicated that women over the age of 25 had the highest proportion of infections (74.6%). Similarly, a study from Addis Ababa, Ethiopia, found that 50.6% of HR HPV-infected women were between the ages of 30 and 50 years [9]. These studies supported our findings that 64.9 percent of HR HPV-infected women were between the ages of 20 and 40, with a significant connection between age group and HR HPV infection (p-value = 0.018).

Furthermore, our investigation identifies the prevalent HR HPV genotype 33, which was found in four LSIL patients and twelve HSIL cases. Other HR HPV genotypes 16, 18 were found in two cases of SCC and adenocarcinoma, respectively. HR HPV genotype 45 was found in four cases of HSIL and 58 in two cases of LSIL. HR HVP genotypes were not found in NILM. This cross-tabulation revealed a statistically significant connection, with a p-value of 0.001.

These findings differ from those reported by Ali et al. [8] in Ethiopia, who discovered that high-risk HPV genotypes were associated with cervical cytology findings. HR HPV 16 was detected in 50% of HSIL reports, while "other HR HPV genotypes" were the most common finding for LSIL, but the results of a study done by Gary C. et al. in his meta-analysis stated that the most frequent genotypes were "other HR HPV" [7].

According to multiple research, HPV-positive women in Sub-Saharan Africa are less likely to be infected with HR HPV 16 than their European counterparts [9]. The overall prevalence of abnormal cytology and positive HR HPV was higher in the 20-40 age group, where women are sexually more active, as evidenced by 16 cases and 12 cases in the 41-80 age group, indicating that abnormal cytology may take years to develop abnormal cervical lesions, which was significantly associated with abnormal cytology.

Furthermore, these findings contrast with those of an Ethiopian investigation, which found that the total prevalence of aberrant cytology was 13.1%. Approximately three-fourths (72%) of HR HPV-infected women were between the ages of 31 and 60 years, which was substantially linked with aberrant cytology [8].

The potential impact of HIV co-infection effects on HPV genotype patterns and cervical lesion progression requires HPV epidemiological data from Sudan, and the absence of documented HIV status represents a notable limitation. HIV immunosuppression status may influence genotype-specific risks or the interplay between HPV and HIV in cervical carcinogenesis.

Conclusion.

The loads of HR HPV infection and cervical cytology abnormalities reported in this investigation are commensurate with the projected prevalence in Sub-Saharan Africa and greater than those reported in Sudan.

Unlike earlier investigations, the high-risk HPV 33" genotype significantly contributed to the overall HR HPV burden. Infections were detected in sexually active women.

Women with cervical cytology abnormalities had the highest HR HPV positive rate. The performance and accuracy of the HPV-DNA PCR and cytology screening methods may need to be validated within run and between using genotype-specific HPV positive control prepared from histologically confirmed positive cervical samples.

Multiplex PCR-based HPV genotypes assay provides essential molecular surveillance for women ≥18 years, detecting high-risk infections during the latent phase before cytomorphological changes manifest. This approach facilitates timely risk stratification when cytological screening yields negative results but viral persistence exists.

Whole-genome amplification (WGA) offers superior sensitivity for broad-spectrum HPV genotyping compared to single-gene PCR, enabling more comprehensive detection of co-infections with multiple HPV genotypes and other sexually transmitted viruses (e.g., HIV, HBV). This approach enhances viral characterization precision while identifying concurrent infections that may influence clinical outcomes.

Furthermore, the screening program for young sexually active women should be promoted in a variety of health care settings. The Ministry of Health should also investigate releasing vaccines that target other carcinogenic HPV types besides genotypes 16 and 18.

A large-scale nation-wide community-based cohort research will also be created and implemented to identify the national burden, and molecular epidemiology of persistent HPV types and cervical cytology abnormalities, allowing us to prescribe the best screening strategy for the local context. These efforts will make a substantial contribution to national cervical cancer prevention strategies.

Additional nationwide research is necessary to examine the prevalence of HPV genotype 33 and the geographic distribution of other high-risk HPV genotypes across different regions of the country. This investigation should particularly assess potential epidemiological changes following the return of refugees from East African countries, as population movements may influence viral genotype circulation patterns.

Such studies would enhance understanding of regional HPV epidemiology and inform targeted prevention strategies. The high prevalence of HPV-33 suggests possible regional viral adaptations, host genetic factors, or synergistic oncogenesis, this supports the multi-genoytpes exposure and mainly co-infections with HPV-16/18.

Furthermore, the findings will help guide the selection of the optimum vaccine formulation based on changes in circulating HPV genotypes to specifically target HPV-33 alongside other high-risk genotypes through nonavalent vaccine implementation in Sudan.

Future research should prioritize combined HPV-HIV screening for those reasons. First, emerging regional HIV trends may substantially impact HPV infection and cervical cancer progression. Second, comparative evidence from other African populations demonstrates distinct HPV genotype profiles between HIV-positive and HIV-negative women. Third, immunodeficiency states are known to accelerate HPV-related oncogenesis, potentially altering observed genotype-disease severity relationships. Implementing standardized HIV testing in subsequent studies would enable more accurate risk stratification and support development of improved cervical cancer prevention strategies for HIV exposed individuals in Sudan.

Therefore, future studies should incorporate simultaneous HPV-HIV screening using validated diagnostic methods, with stratified analysis by HIV status to elucidate genotype-specific oncogenic risks accompanied with regular CD4+ monitoring should be implemented to evaluate immunological modifiers for HIV-positive participants.

Conflicts of Interest: All authors declared no conflict of interest.

REFERENCES

1. World Health Organization. Cervical cancer. 2024. https://www.who.int/news-room/fact-sheets/detail/cervical-cancer
2. World Cancer Research Fund. Cervical cancer statistics. 2024. https://www.wcrf.org/cancer-trends/cervical-cancer-statistics
3. ICO/IARC HPV Information Centre. Sudan: Human papillomavirus and related cancers, fact sheet. 2023. https://hpvcentre.net/statistics/reports/SDN_FS.pdf
4. International Agency for Research on Cancer. Cervical cancer global statistics. 2024. https://www.iarc.who.int
5. Omire A, Budambula N. L. M, Kirumbi L, et al. Cervical dysplasia, infection, and phylogeny of human papillomavirus in HIV-infected and HIV-uninfected women at a reproductive health clinic in Nairobi, Kenya. BioMed Research International. 2020:4945608.
6. Travill D.I, Machalek D.A, Rees H, et al. High prevalence of human papillomavirus in unvaccinated adolescent girls in South Africa, particularly those living with HIV. Vaccine. 2024:42.
7. Mohan L, Karthika M. A community-based cervical cancer screening program in Alappuzha district of Kerala using camp approach. Journal of Family Medicine and Primary Care. 2022;11:1152-1157.
8. Ali K.E, Mohammed I.A, Difabachew M.N, et al. Burden and genotype distribution of high-risk human papillomavirus infection and cervical cytology abnormalities at selected obstetrics and gynecology clinics of Addis Ababa, Ethiopia. BMC Cancer. 2019;19:768.
9. Wong L.P, Wong Y.L, Low W.Y, et al. Cervical cancer screening attitudes and beliefs of Malaysian women who have never had a Pap smear: A qualitative study. International Journal of Behavioral Medicine. 2008;15:289-292.
10. Ali K.E, Mohammed I.A, Difabachew M.N, et al. Burden and genotype distribution of high-risk human papillomavirus infection and cervical cytology abnormalities at selected obstetrics and gynecology clinics of Addis Ababa, Ethiopia. BMC Cancer. 2019;19:768.
11. Kim T.E, Kim H.W, Lee K.E. Distribution of human papillomavirus 52 and 58 genotypes, and their expression of p16 and p53 in cervical neoplasia. Korean Journal of Pathology. 2014;48:24.