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CHALLENGES OF FALSE POSITIVE CYTOLOGICAL RESULTS IN CERVICAL CANCER SCREENING: THE FUTURE PERSPECTIVE OF MORE RELIABLE TESTING METHODS

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Summary

Introduction: Cervical dysplasia caused by human papillomavirus is a significant public health concern that might lead to cervical cancer, particularly in the developing world. The cervical cancer preventative screening program in Kazakhstan needs more sophisticated screening methods for improving early detection and management, which can ultimately reduce the burden of this disease on women's health.

Aim: The aim of this study was to elicit false positive results as a major shortcoming of a current screening method and to recommend more accurate testing algorithms.

Materials and Methods: Study design: Modelling study, encompassing a period between 2016-2023 in The Medical Centre Hospital of President's Affairs Administration of the Republic of Kazakhstan, Astana, Kazakhstan. The diagnoses of cervical dysplasia based upon cytological and human papillomavirus (HPV) DNA tests were collected in a hospital database (Infomed) and further analysed through GraphPad software.

Results and discussions: HPV-negative tests occurred in 30.43% (2016), 45.45% (2017), 28.20% (2018), 42.5% (2019), 35.71% (2020), 32.14% (2021), 21.15% (2022), and 54.45% (2023) of the total amount of patients. Nevertheless, they were diagnosed with mild dysplasia. While screening saves lives, it's important to acknowledge that false positives are inevitable without optimisations, and excessive testing can lead to substantial harm, including unnecessary treatments and a burden on the healthcare system.

Conclusion: Improvements in specificity might lead to a reduction in false positive results and an increase in the accuracy of referrals for colposcopy. The utilization of dual-staining with p16/Ki-67 cytology appears promising as a biomarker-based method for determining further steps in cervical cancer screening.

Keywords: *Cervical dysplasia, cervical cancer screening, human papillomavirus, HPV genotype, p16/Ki-67 cytology*

Резюме

ВЫЗОВЫ ЛОЖНОПОЛОЖИТЕЛЬНЫХ ЦИТОЛОГИЧЕСКИХ РЕЗУЛЬТАТОВ В СКРИНИНГЕ РАКА ШЕЙКИ МАТКИ: ПЕРСПЕКТИВЫ БОЛЕЕ НАДЕЖНЫХ МЕТОДОВ ТЕСТИРОВАНИЯ

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Введение: Дисплазия шейки матки, вызванная вирусом папилломы человека, представляет собой значительную проблему для общественного здоровья, которая может привести к раку шейки матки, особенно в развивающихся странах. Программа скрининга для предотвращения рака шейки матки в Казахстане требует более совершенных методов скрининга для улучшения ранней диагностики и управления, что в итоге может снизить бремя этого заболевания для здоровья женщин.

Цель: целью данного исследования было выявление ложноположительных результатов как основного недостатка текущего метода скрининга и рекомендация более точных алгоритмов тестирования.

Материалы и методы: Дизайн исследования: Моделированное исследование, охватывающее период с 2016 по 2023 год в Больнице Медицинского центра Управления делами Президента Республики Казахстан, Астана, Казахстан. Диагноз дисплазии шейки матки, основанный на цитологических и молекулярно-генетических тестах на вирус папилломы человека (HPV), был собран в базе данных больницы (InfoMed) и дополнительно проанализирован с использованием программного обеспечения GraphPad.

Результаты и обсуждение: Отрицательные результаты HPV выявлены у 30,43% (2016), 45,45% (2017), 28,20% (2018), 42,5% (2019), 35,71% (2020), 32,14% (2021), 21,15% (2022), 54,45% (2023) от общего числа пациентов. Тем не менее, у них была диагностирована легкая дисплазия. Несмотря на то, что скрининг спасает жизни, важно признать, что ложноположительные результаты неизбежны без оптимизации, и избыточное тестирование может привести к значительному вреду, включая необоснованные лечения и нагрузку на систему здравоохранения.

Вывод: Улучшение специфичности может привести к уменьшению ложноположительных результатов и повышению точности направлений на колпоскопию. Использование двойной окраски с цитологией p16/Ki-67 кажется перспективным методом на основе биомаркеров для определения дальнейших шагов в скрининге рака шейки матки.

***Ключевые слова**: дисплазия шейки матки, скрининг рака шейки матки, вирус папилломы человека, генотип ВПЧ, цитология p16/Ki-67.*

Түйіндеме

ЖАТЫР МОЙНЫ ОБЫРЫНЫҢ СКРИНИНГІНДЕГІ ЖАЛҒАН ПОЗИТИВТІ ЦИТОЛОГИЯ НӘТИЖЕЛЕРІНІҢ ҚИЫНДЫҚТАРЫ: СЕНІМДІРЕК ТЕСТІЛЕУ ӘДІСТЕРІНІҢ ПЕРСПЕКТИВАЛАРЫ

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Кіріспе: Дамушы мемлекеттерде адам папилломавирусынан туындаған жатыр мойынының дисплазиясы жатыр мойны обырына әкелуі мүмкін денсаулық сақтаудың маңызды проблемасы болып табылады. Қазақстандағы жатыр мойны обырының алдын алу бойынша скринингтік бағдарлама ерте диагностика мен басқаруды жақсарту үшін скринингтік тексеру әдістерін жетілдіруді талап етеді.

Материалдар мен тәсілдер. Зерттеу дизайны: Қазақстан Республикасы Президенті Әкімшілігінің Медициналық орталығының ауруханасында 2016 жылдан 2023 жылға дейінгі кезеңді қамтитын модельдік зерттеу, Астана, Қазақстан. Бұл зерттеудің **мақсаты** қазіргі скрининг әдісінің негізгі кемшілігі ретінде жалған-оң нәтижелерді анықтау және дәлірек тестілеу алгоритмдерін ұсыну болып табылады.

Әдістері: Жатыр мойны дисплазиясының диагностикасы адам папилломавирусына (HPV) цитологиялық және молекулярлық-генетикалық сынақтар негізінде ауруханының дерекқорынан (InfoMed) жиналды және GraphPad бағдарламалық құралының көмегімен әрі қарай талданды.

Нәтижелер және талқылау: HPV теріс нәтижелері науқастардың жалпы санынан 30,43% (2016), 45,45% (2017), 28,20% (2018), 42,5% (2019), 35,71% (2020), 32 ,14% (2021), 211% (2022), 54,45% (2023) екені анықталды. Алайда оларға жеңіл дисплазия диагнозы қойылды. Скрининг өмірді сақтап қалса да, оңтайландырусыз жалған оң нәтижелер болмай қоймайтынын мойындау маңызды, ал артық тестілеу денсаулық сақтау жүйесіне артық ауыртпалықты қоса, қажетсіз ем елеулі зиянға әкелуі мүмкін.

Қорытынды: Жақсартылған спецификалық жалған-оң нәтижелердің төмендеуіне және кольпоскопияға сілтемелердің дәлдігінің жоғарылауына әкелуі мүмкін. p16/Ki-67 цитологиясымен қосарланған бояуды пайдалану жатыр мойны обырын скринингтің келесі қадамдарын бағыттау үшін биомаркер негізіндегі перспективалы әдіс болып көрінеді.

***Негізгі сөздер:** жатыр мойны дисплазиясы, жатыр мойны обырының скринингі, адам папилломавирусы, HPV генотипі, p16/Ki-67 цитологиясы.*

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Introduction

Most human papillomavirus (HPV) infections are temporary and become undetectable within 12 to 24 months (10). However, in some women where the infections persist, there is a significant risk of developing precancerous conditions. Numerous studies have affirmed that persistent infection with an oncogenic HPV type is the primary risk factor for the development of cervical intraepithelial neoplasia (CIN), which can vary in severity from CIN1 to CIN3, and ultimately, cancer (25, 18, 2, 14).

The natural progression of CIN lesions differs based on their grade. CIN1 is categorized as a low-grade squamous intraepithelial lesion (LSIL). Statistical data indicates that over 70–80% of CIN1 lesions either spontaneously regress without treatment or become undetectable (10, 15). Consequently, CIN1 is often considered more of a state of infection than an advanced stage of disease. Detecting CIN1 following an HPV infection does not necessarily signify disease progression, and observed clearance rates may be due to the inability to detect the infection (18). Therefore, caution is advised when interpreting clearance rates.

In contrast, CIN2 and CIN3 are designated as high-grade dysplasia or high-grade squamous intraepithelial lesions (HSIL). CIN2 exhibits two distinct pathways: the annual regression rate for CIN2 in adult women is estimated to fall between 15% and 23%, with up to 55% experiencing regression within 4–6 years (15, 21). Approximately 2% of CIN2 lesions advance to CIN3 within the same timeframe. CIN3 is recognized as a genuine precancerous stage with the potential to progress to invasive cancer, occurring at a rate of 0.2% to 4% within 12 months (15, 7).

Untreated CIN3 carries a 30% probability of evolving into invasive cancer over a span of 30 years. However, when appropriately treated, only about 1% of CIN3 cases progress to invasive cancer (15, 7, 25). Adenocarcinoma of the cervix, distinct from squamous cell carcinoma, originates from the glandular epithelium of the endocervical canal, with its immediate precursor being adenocarcinoma in situ (19).

The introduction of organized cervical cancer screening programs has played a role in reducing cervical cancer incidence and mortality in various Western countries. The program to reduce mortality from cervical cancer in Kazakhstan is based on three main principles (1):

1. Primary Prevention: Promoting a healthy lifestyle and eliminating potential risk factors, as well as vaccinating specific population groups.
2. Secondary Prevention: Implementing high-quality and well-organized screening for women in the population.
3. Standardizing the diagnosis and treatment protocols for invasive cervical cancer and providing palliative care, following uniform guidelines for all.

The primary screening involves collecting material for cytological examination from the cervix –conventional and liquid-based cytology using the Papanicolaou method (Pap test), performed once every 5 years (1).

Medical Centre Hospital of the President's Affairs Administration of the Republic of Kazakhstan is one of the leading hospitals in Kazakhstan with its state-of-the-art technology and high-profile health professionals. Such a combination can guarantee that the hospital is among the first adapters of national screening programs while many other hospitals throughout Kazakhstan might not be as successful due to the lack of health professionals, finances, facilities, and laboratories. The hospital can serve as a marker for how national screening programs to fight cervical cancer are being implemented and what results are being acquired.

Based on a database of patients who have gone through cytology and HPV co-testing screening from 2016 and 2023, it has been observed an increasing number of women with abnormal Pap smears (cervical dysplasia), – from ASCUS (Atypical Squamous Cells of Undetermined Significance) to LSIL or HSIL with or without positive HPV. This raises the question of the possibility of false positive results, as cervical dysplasia cannot appear without the existence of HPV in the body.

This study aimed to elicit the possibility of a false positive screening result within an organized cervical cancer screening program due to the design of the screening program itself, – taking cytology tests (Pap test) as a primary testing method instead of HPV molecular test as a primary testing step during screenings. The relevance of our report might be taken into consideration in order to reduce over-treatment, patient anxiety and adherence to multiple screenings due to false positive results, and an overall reduction of healthcare system burden.

Methods

Cytological and PCR testing

Samples from the cervix are collected by experienced gynecologists using FLOQSwabs in eNAT medium (Copan, Besozzo, Italy) for the detection of HPV DNA.

Samples for the Thin Prep Pap test were obtained by inserting a cytobrush into the endocervical canal, which were then immediately placed in containers with Thin Prep transport medium containing methanol. The samples were stored at a temperature of 15 to 20°C and delivered to the laboratory within 4 hours of collection.

On the Thin Prep 2000 processor (Cytyc Corporation, USA), preparations were made and stained with the Papanicolaou stain. Cervical cytology samples were classified based on the Bethesda System terminology (TBS): Atypical Squamous Cells of Undetermined Significance (ASCUS), Low-Grade Squamous Intraepithelial Lesion (L-SIL), High-Grade Squamous Intraepithelial Lesion (H-SIL). The histological grading of the severity of the lesion depends on the quantity and location of immature undifferentiated cells within the epithelial layer - its stratification. CIN I - undifferentiated cells occupy the lower third of the epithelial layer. CIN II - immature cells occupy the lower two-thirds of the epithelial thickness; CIN III (including severe dysplasia and preinvasive cancer) - abnormal immature cells occupy more than two-thirds of the epithelial thickness or the entire thickness, but there is no invasion into the underlying stroma.

HPV PCR DNA testing

Since 2015, the Hospital has been using PCR test kits with amplifiers "iQ5 iCycler," "CFX96," and the "RealBest Diagnostics" program for HPV PCR testing. A segment of the purified DNA initially used for HPV PCR detection, underwent genotyping. Subsequently, 10 μL of template DNA was subjected to amplification. This program automates all the operations for analysis, rejection, and result calculations. Samples were processed on the DX-Real-Time System PCR analyzer.

Diagnosis

There are varying degrees of dysplasia in cervical lesions. According to the Lower Anogenital Squamous Terminology (LAST) project, pathologists categorized these specimens as either low-grade squamous intraepithelial lesions (LSIL) or high-grade squamous intraepithelial lesions (HSIL). LSIL, which was traditionally known as CIN1 or mild dysplasia, typically does not necessitate immediate treatment and should be managed conservatively. Patients diagnosed with LSIL are typically advised to return for follow-up in one year, which involves a repeat Pap smear along with HPV molecular testing. This approach is taken because a majority of LSIL lesions tend to regress on their own.

In contrast, HSIL, traditionally referred to as CIN2/3 or CIS typically requires a more proactive approach to treatment. Patients diagnosed with HSIL often undergo an excisional procedure to address the condition effectively.

Data sourcing

The Medical Center Hospital of the President's Affairs Administration of the Republic of Kazakhstan (The Hospital) has a well-organized cohort of women over the age of 18, with a population of over 10,000 individuals. Since 2016, as part of the mandatory preventive examination, all women have undergone gynecological examinations, including liquid-based cytological analysis of cervical smears stained with the Papanicolaou method. In case of atypical results, PCR detection and quantitative determination of DNA from high-risk human papillomaviruses 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, and 59 genotypes are conducted, and if the test is positive, colposcopy is performed. The hospital maintains control over timely screening, operates a notification and tracking system for abnormal results, and actively invites individuals for screening, making screening a top priority. The team of specialists is highly qualified at all stages of screening.

The study involves a retrospective analysis of real-time PCR and liquid cytology findings obtained in the Hospital laboratory during the periods of December 2016, 2017, 2018, 2019, 2020, 2021, and 2022. The examined results were obtained from women in the Kazakh population aged 18 and above who visited the hospital for routine cervical cancer screening. Results from women with a known diagnosis of cervical cancer were included in a separate group.

Data Sources:

"Report on Malignant Diseases" (form No. 7) for the Republic of Kazakhstan for the years 2016 and 2022.

Data from the National Center for Healthy Lifestyle Formation on the results of screening examinations of target population groups in Kazakhstan for the years 2016 and 2022.

Data from the hospital's laboratory service on the results of cytological examination of cervical smears stained with the Papanicolaou method and real-time PCR for the detection of high-risk human papillomavirus DNA (12 genotypes).

Data from the hospital's "InfoMed" information database: demographic, clinical, and laboratory data.

Ethics

The study was conducted in accordance with the Helsinki Declaration and was approved by the Local Ethics Committee (Protocol No. 3, dated 9.08.2022). Informed consent from patients was not required for this study because all the data used were obtained as part of routine comprehensive preventive examinations from the laboratory without the inclusion of personal data. Furthermore, each patient at the hospital provides consent for their data to be entered into the hospital's information system, "InfoMed" and for their data to be used in scientific research.

Results and Discussion

In 2016 and 2017, 23 and 22 women (ages ranging between 25 (min) and 69 (max), and an average being 42.89 and 42.36 years old, respectively) were diagnosed with cervical neoplasia. 9 women in 2016 were diagnosed with less aggressive mild dysplasia (CIN1) while the rest were diagnosed with severe dysplasia (CIN2 and CIN3) (n=14), which is almost inversely to 2017 where the majority were diagnosed with mild dysplasia (n=16). However, HPV testing performed on all women elicited negative results in 7 out of 23 women in 2016, and 10 out of 22 women in 2017.

In 2018, 39 women were diagnosed with cervical dysplasia, the youngest being 23 and the oldest 71 years old, with the average being 40.89 years old. Cytological examination of cervical smears stained with Papanicolaou staining showed that 2 patients were displaying abnormal cytologic changes that are suggestive of dysplasia, 26 patients with mild dysplasia, and 11 with severe dysplasia. Contrastingly, PCR tests showed no HPV in 17 women.

Similarly, 40 and 42 women were diagnosed with cervical dysplasia in 2019 and 2020, respectively. The average age for 2019 was 41.42 (min/max=22/68), and 37.35 (min/max=23/68) for 2020. But during performing subsequent tests for HPVs, 17 out of 40 women in 2019 and 15 out of 42 women in 2020 showed negative results. Nonetheless, 25 and 33 women were diagnosed with CIN1 in 2019 and 2020, respectively, while others were diagnosed with CIN2-3.

2021 wasn't much of an exception among other years, with 28 women with ages ranging between 23 and 62 (average=41.42) diagnosed with cervical dysplasia, and 25 of them were diagnosed with mild dysplasia, though 9 patients out of total tested negative for HPVs.

In 2022 and 2023 the respective 52 and 101 women were registered with cervical neoplasia (the vast majority were diagnosed with CIN1 (47 in 2022, 98 in 2023)). The average age for both years was 38 years, (min/max=24/73 and =21/59 in 2022 and 2023, respectively). 11 women out of 52 tested negatives for the HPV PCR test in 2022, and a half (55 out of 101) tested negative in 2023.

The average age range of women diagnosed with cervical dysplasia between 2016 and 2023 is on par with the global statistical results, – for instance, annually approximately 250,000 to 1 million women aged between 25 and 35 years in the United States receive a diagnosis of cervical dysplasia (5).

Cervical neoplasia is the term used to describe the abnormal growth of cells in the cervix, the lower part of the uterus that connects to the vagina. Numerous studies have consistently affirmed that persistent infection with high-risk types of human papillomavirus (HPV) is the primary contributing factor to the development of cervical intraepithelial neoplasia (CIN), which encompasses CIN1 to CIN3 and the potential for cancer progression (18, 25, 14). Specifically, the VIVIANE study has identified HPV33 and HPV16 as having the strongest associations with the risk of CIN development, followed by HPV18, HPV31, and HPV45 (18). Moreover, it is supported by other statistical analyses which concluded that HPVs are responsible for the development of 99.7% of cervical cancer in women as a result of untreated cervical dysplasia (13, 17). All these findings state that HPV is essential for the development of cervical dysplasia. However, as shown from our results, HPV-negative tests occurred in 30.43% (2016), 45.45% (2017), 28.20% (2018), 42.5% (2019), 35.71% (2020), 32.14% (2021), 21.15% (2022), 54.45% (2023) of the total amount of patients diagnosed with cervical dysplasia. This raises a concern revolving around the significant number of false-positive results for women who participate in cervical screening programs in the hospital. Screenings for women for gynaecological abnormalities first involve liquid-based cytology (LBC), and if abnormalities are found then molecular testing for HPV infection is performed. However, HPV DNA testing as a screening tool has already proven to be more effective in detecting cervical dysplasia than cytology tests (20). Our results can be also supported by the recent study that stated primary LBC screening approach results in approximately 95 false-positive results, with a confidence interval of 93% to 97% (8). Additionally, this screening misses approximately 4.9 cases of CIN grade 2/3 per 1000 women (95% Confidence Interval (CI) ranging from 3.5 to 6.7).

Screening tests themselves, while valuable, are not without their limitations when it comes to adequately identifying CIN. According to figures from a Koliopoulos review (12), out of every 1000 women screened, 20 will be diagnosed with CIN of grades 2 or 3 (CIN2/3). Cytology alone can detect 15 of these women, while screening with HPV testing would identify an additional 3 who might have been missed with cytology. This heightened sensitivity of HPV testing, however, does come at the cost of reduced specificity compared to LBC.

The increased confidence in primary HPV testing, as opposed to undergoing cytology testing alone (performed at the same frequency), is due to the heightened sensitivity of the HPV test (22, 9). It correlates with the Cochrane database of systematic reviews that concluded a negative HPV test could be more reassuring (12). Recent multicentre studies have provided evidence that HPV-based screening offers greater protection against invasive cervical carcinomas compared to cytology-based screening (11, 20). In these studies, the recorded cumulative incidence of cervical cancer was lower than 5.5 years after receiving a negative HPV test result compared to 3.5 years after a negative cytology result. This empirical evidence suggests that 5-year screening intervals for HPV are safer and more effective than the 3-year intervals typically used for cytology-based screening.

As a result, Western Countries have already implemented primary HPV screening in their national programs (23,24). An updated cervical cancer screening guideline from the American Cancer Society (ACS) recommend starting screening at age 25 with an HPV test and retesting every 5 years till the age of 65. However, co-testing with an HPV/Pap every 5 years or with a Pap test every 3 years still holds its relevance (6).

However, it's important to note that if primary HPV screening is not implemented optimally, it can lead to a higher number of referrals for colposcopy (8). It becomes crucial to establish what level of false positive is deemed acceptable within our screening programs. Cervical screening undoubtedly saves lives, but it must be applied thoughtfully to achieve its maximum benefits while minimizing the consequences of over-treatment in cases of false positives. Moreover, the choice of which screening method to employ is not solely a scientific matter but also a question of effectively allocating scarce public health resources, which is apparent in Kazakhstan.

There are still major limitations of HPV testing as a primary testing. While this capability is clinically interesting and potentially valuable, it may not always be clinically relevant, especially considering that most HPV infections (about 80%) will naturally clear within two years. Furthermore, the lifetime risk of encountering HPV is quite high, estimated at around 90%.

Given the inability to predict which infections will persist, the focus lies on detecting persistent infections. Women who have a persistent infection are at an elevated risk of developing cervical neoplasia. Typically, in screening programs, women with an ASCUS cytology result are called back for repeat cytology after six months if HPV testing is not performed. Meanwhile, women with an LSIL result are referred for colposcopy-directed biopsy.

Referring all women who test positive for HPV or have an LSIL smear result for additional investigations, such as repeat smears, colposcopy, and biopsy, could potentially increase the sensitivity of cancer screening. However, it also comes with the risk of increasing morbidity. Additionally, the prospect of repeat tests may discourage women from participating in screening, potentially leading to reduced screening rates. This underscores the need to strike a careful balance between enhancing sensitivity and

minimizing the potential harms and drawbacks of over-testing and over diagnosis.

In cervical cancer screening, it's not only important to increase sensitivity but also specificity to effectively detect cancers and precancers while reducing overall morbidity. To achieve this, there's a need to triage HPV-positive women or women with slightly abnormal smear results (LSIL) to reduce the number of false positives.

One potential triage system for these cases could involve additional staining with p16/Ki-67. p16 is a protein involved in regulating the cell cycle, typically causing cell cycle arrest under normal conditions (4, 27). Ki-67, on the other hand, is a marker for cell proliferation. In normal circumstances, p16 and Ki-67 are seldom seen together. However, the overexpression of both p16 and Ki-67 suggests a cell cycle deregulation due to HPV and indicates a high-grade lesion (28, 16, 3). This concept of dual-stain cytology, often referred to as the CINtec1 PLUS Cytology test, represents a morphology-independent biomarker approach. It could be more accurately described as "diagnostic" cytology, offering a promising approach to improving specificity in cervical cancer screening. In a screening population in Belgium covering individuals aged 25 to 65 years, dual-stain cytology exhibited significantly higher sensitivity at 66% while maintaining a slightly lower specificity at 1.0% (26). Improved sensitivity means that less disease is missed, while enhanced specificity means fewer false-positive results. The latter aspect provides the potential to reduce morbidity by minimizing the need for additional examinations, such as repeat cytology, colposcopy, and biopsies. Additionally, this can lead to a decrease in the number of women who discontinue their participation in cervical cancer screening programs.

Conclusion and Future Perspectives

Cervical neoplasia poses a significant concern in women's health, as it has the potential to progress into full-blown cervical cancer if not addressed. Timely detection and appropriate treatment can effectively prevent the development of invasive cervical cancer and greatly improve women's overall health. Thus, it is crucial for women to undergo regular cervical cancer screening to detect and manage cervical neoplasia at an early stage using Pap smears or HPV testing. However, our study reports that cytology tests might result in false positive results, as HPV tests show negative results of HPV. Cervical screening, regardless of the testing method used, inherently carries a degree of uncertainty. While this study provides valuable insights into optimal screening strategies, it's essential to acknowledge that screening, while lifesaving, cannot attain perfection.

There exists an inherent trade-off in strategies aimed at increasing detection rates. These strategies often lead to a disproportionate increase in false positives, resulting in unnecessary over-treatment. This trade-off becomes particularly relevant in Kazakhstan, where most hospitals are under-financed, and under-equipped with sophisticated lab facilities, up-to-date equipment and testing tools, and healthcare professionals. Such a requirement is unattainable, and as our report illustrates, striving to approach this standard might do more harm than good. The current approach of doing the PEP smear test as a primary step for screening is not conducive to public health, sustainability, or fostering a clear understanding of screening's capabilities and limitations among the public. Screening is a critical endeavor for reducing cervical cancer mortality, and its strengths and limitations must be viewed in context to maximize its benefits.

The findings of this analysis should prove valuable in optimizing screening approaches and highlighting the complexities of different implementations, allowing for informed decision-making.

Another effective preventive measure against cervical neoplasia is HPV vaccination, as it can provide protection against high-risk HPV types that are linked to the development of cervical abnormalities and cancer. Looking ahead to the future of cervical screening, it's crucial to consider the remarkable international success of the HPV vaccine. Countries with high rates of HPV vaccine uptake are already witnessing a significant decline in rates of precancerous and abnormal cervical cells.

While screening is an extraordinary lifesaving measure, it's equally important to comprehend its inherent limitations and intricacies. This understanding ensures that national programs yield the maximum benefit while minimizing misunderstandings.

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