Arh. farm. 2024; 74: 56 – 75 *Review article/Pregledni rad*

Human papillomaviruses and cervical cancer from the perspective of the World Health Organisation initiative for cervical cancer elimination

Brankica Filipić[1]*, Ivana Rapajić-Moran[2], Ines Nikolić[3,4], Slavica Oljačić[5], Aljoša Mandić[6,7]

[1]Department of Microbiology and Immunology, University of Belgrade – Faculty of Pharmacy, Vojvode Stepe 450, 11221 Belgrade, Serbia

[2]Department of Social Pharmacy and Pharmaceutical Legislation, doctoral studies, University of Belgrade – Faculty of Pharmacy, Vojvode Stepe 450, 11221 Belgrade, Serbia

[3]Department of Pharmaceutical Technology and Cosmetology, University of Belgrade – Faculty of Pharmacy, Vojvode Stepe 450, 11221 Belgrade, Serbia

[4]School of Pharmaceutical Sciences, University of Geneva – Faculty of Science

[5]Department of Pharmaceutical Chemistry, University of Belgrade – Faculty of Pharmacy, Vojvode Stepe 450, 11221 Belgrade, Serbia

[6]Department of Gynecologic Oncology, Oncology Institute of Vojvodina, Put dr Goldmana 4, 21204 Sremska Kamenica, Serbia

[7]Department of Gynaecology and Obstetrics, University of Novi Sad – Faculty of Medicine, Hajduk Veljkova 3, 21137 Novi Sad, Serbia

*Corresponding author: Brankica Filipić, e-mail: brankica.filipic@pharmacy.bg.ac.rs

Abstract

Human papillomaviruses (HPV) are the most common sexually transmitted pathogens worldwide, leading to infections with a wide range of clinical manifestations: from benign conditions to different types of cancer in women and men as well. Cervical cancer is highly correlated with persistent high-risk-HPV (HR-HPV) infection, which is the key factor in emergence of 99.99% of cervical cancer cases. The most effective way to prevent HPV-related cancers is vaccination. There are three available prophylactic HPV vaccines: bivalent, quadrivalent and nonavalent. The nonavalent vaccine is gradually replacing other HPV vaccines in most countries and can be given from year 9, but it is commonly routinely implemented at the

age of 11 to 12. The World Health Organization has recognised cervical cancer as a global threat and has announced the so-called 90-70-90 strategy to reduce and even eliminate cervical cancer. This strategy implies that 90% of girls should be vaccinated by the age of 15, 70% of women should be screened for cervical cancer, and 90% of women diagnosed with cervical disease should receive adequate treatment. Although different treatment options are available: surgery, radiation therapy, chemotherapy, and advanced target therapy using monoclonal antibodies, great efforts are needed to achieve the goals set by the World Health Organization to eliminate cervical cancer.

Key words: high-risk HPV (HR-HPV) infection, cervical cancer, HPV vaccination

---

doi.org/10.5937/arhfarm74-48226

Introduction

Human papillomaviruses (HPV) are small (52-55 nm), icosahedrally symmetric, non-enveloped viruses with circular double strand DNA placed within the capsid made of 72 pentameric capsomers and two structural proteins: L1 (major capsid protein) and L2 (minor capsid protein). HPV are strict human pathogens belonging to the *Papillomaviridae* family, with the affinity to infect cutaneous and mucosal epithelial tissues (1).

More than 200 strains of HPV have been identified, of which approximately 40 types infect the anogenital region (2). Between 5 and 18 of these HPV strains have been classified as high-risk (HR) genotypes. HPV are the most common sexually transmitted pathogens worldwide, with clinical manifestations spanning from benign to malignant processes in women and men. The International Agency for Research on Cancer has recently classified 12 HPV genotypes as carcinogenic, including 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, and 59 (group 1). HPV 68 is also considered probably carcinogenic or group 2a. Additionally, HPV genotypes 26, 30, 34, 53, 66, 67, 69, 70, 73, 82, 85, and 97 have been linked to rare causes of cervical cancer and are classified as possibly carcinogenic (group 2b). Globally, HPV 16 and HPV 18 are responsible for approximately 70% of cervical cancer cases (3).

The virus enters the body via cutaneous or mucosal microlesions and around 85% of the sexually active population (regardless of their sex, gender identity, or sexual orientation) have been infected with HPV during their lifespan. In a great majority of cases, HPV infection is asymptomatic, with 70% of the infected clearing the infection in one year, and 91% clearing the infection in two years (4).

The most common clinical presentations of cutaneous HPV infection are anogenital warts that can appear on the vagina, penis or anus, and are highly associated with low-risk genotypes HPV 6 and HPV 11, which cause 90% of genital warts (5). Genital warts usually cause emotional discomfort for the patient, and the treatment can be long and painful, with frequent recurrent infections.

On the other hand, HPV causes about 5% of cancers worldwide, and it is estimated that 625,600 women and 69,400 men get HPV-related cancer each year (6).

When HPV infection caused by HR oncogenic genotypes is not well controlled by the immune system, it can lead to persistent infection for many years and, if untreated, may cause the development of precancerous and cancerous lesions. HPV infection can cause six types of cancers: it is related to 99.99% cervical cancers, 90% of anal cancers, 50% of penile cancers, 70% of vaginal and vulvar cancers, and 20-60% of oropharyngeal cancers (7, 8). Although early detection of HPV-related cancers can be achieved with appropriate screening methods, the frequency of HPV-related disease is particularly high in developing countries. As the most common HPV-related cancer is cervical cancer, the aim of this paper is to analyse, describe and present the available options for prevention and treatment of this type of cancer. Special attention will be directed towards HPV vaccination as the most effective tool for cervical cancer prevention.

HPV and cervical cancer

Cervical cancer is the most common HPV-related cancer, accounting for 3.1% of all cancers worldwide, and the leading cause of cancer deaths in women in the developing world (with 341,831 deaths annually) (8). It is estimated that the global incidence rate of cervical cancer was 13.3 cases per 100,000 women in 2020, with a mortality rate of 7.2 per 100,000 women (9). The incidence of cervical cancer and mortality rate in 2020 within Europe is presented in Table I (10, 11). Montenegro has the highest incidence and death rate from cervical cancer in the European region, while among the European Union countries the highest incidence and mortality rates have been recorded in Romania, Bulgaria, Estonia, Latvia and Slovakia, respectively (Table I). Serbia is ranked as the fifth country in the European region and the second country in the Balkan region by the incidence and death rate from cervical cancer, according to the available data (Table I). Persistent HPV infection is an essential factor that leads to the development of cervical cancer, which is associated with certain HR types of HPV: genotype HPV 16 is responsible for 55-60% of all cervical carcinomas worldwide, while genotype HPV 18 is recognised to cause around 20% of cervical adenocarcinomas (2, 12). Other oncogenic HPV genotypes which can cause about 25% of cervical carcinomas are 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 68 (13). In Serbia, 12 HR-HPV genotypes have been identified as dominant: 16, 18, 31, 33, 35, 45, 52, 53, 58, 59, 62, 66, with the most frequent genotype being 16, followed by 45, and 31 (14). Persistent infection with oncogenic types is related to the development of premalignant changes or dysplasia of squamous cells in the cervical epithelium (also known as *cervical intraepithelial neoplasia* - CIN). Further on, CIN can slowly progress, forming dysplastic structures that are classified into 3 degrees according to their severity: CIN1 (mild), CIN2 (moderate), CIN3 (severe dysplasia) and, finally, as carcinoma *in situ* and invasive carcinoma. CIN1 denotes mild dysplasia where one-third of the lower epithelium shows dysplasia, CIN2 (or moderate dysplasia) represents a condition where two-thirds of the epithelium is affected, and CIN3 (severe dysplasia) denotes a condition where more than two-thirds of the epithelium is affected (12). The process of HR-HPV carcinogenesis starts with HPV entering microlesions and its integration into the host cell genome. HPV self-replicates, spreads to other epithelial cells, causes irregular growth of infected cells and, subsequently, the virions are released through desquamated cells of the cervical epithelium, enabling further transmission (15). Cervical cancer can be detected by HR-HPV testing, by collecting Pap smears and performing colposcopy (16).

Table I Cervical Cancer incidence and mortality rate across Europe (estimates for 2020) (10, 11)

Tabela I Incidenca karcinoma grlića materice i stopa smrtnosti u zemljama Evrope (procene za 2020. godinu) (10, 11)

Country	Cervical cancer incidence[1]*	Number of new cases in 2020, all ages[2]	Cervical cancer mortality[1]*	Number deaths in 2020, all ages[2]
Montenegro	35.6	113	17	54
Romania	34.2	3 380	18.3	1 805
Bulgaria	28.2	1 009	14.1	503
Estonia	28.1	196	8.88	62
Serbia	27	1 205	14.2	634
Latvia	26.3	267	13.4	136
Slovakia	24.9	698	10.1	284
Hungary	24.7	1 251	9.52	482
Republic of Moldova	22.8	480	11.8	248
Ukraine	20.3	4 756	8.90	2 089
Poland	19.8	3 862	11	2 137
Russian Federation	19.6	15 308	9.64	7 550
Bosnia and Herzegovina	18.6	312	9.14	153
Belarus	16.5	835	7.09	358
Portugal	16.1	865	7.05	379
Croatia	15.8	336	7.06	150
Norway	14.8	397	3.58	96
Czechia	14.1	813	7.32	435
Ireland	13.8	342	4.26	106
Denmark	13.2	384	4.81	140
Greece	13.1	697	5.31	282
Sweden	13	656	3.97	200
Germany	11	4 666	4.90	2 075
North Macedonia	10.9	113	5.95	62
Italy	10.2	3 152	3.26	1 011
France	10	3 379	4.31	1 452

Slovenia	9.96	104	5.17	54
Iceland	9.42	16	2.94	5
Albania	9.41	133	5.24	74
Luxembourg	7.76	24	3.23	10
Austria	8.43	385	3.72	170
Spain	8.23	1 957	3.42	814
Cyprus	7.62	46	5.46	33
Malta	5.91	13	2.27	5

*Per 100.000 women;
[1]Source: https://hpvcentre.net/statistics/
[2]Source: GlobalCan https://gco.iarc.fr

Prophylactic HPV vaccines

The most (cost)-effective way to prevent HPV-related cancers and infections is vaccination.

There are three recombinant HPV prophylactic vaccines available (Table II):

- quadrivalent HPV vaccine Gardasil®4 approved by the *FDA* and *EMA* in 2006;
- bivalent vaccine Cervarix™ approved in 2007 (by the *EMA*) and 2009 (by the *FDA*) and
- nonavalent vaccine Gardasil®9 approved in 2014 (by the *FDA*) and 2015 (by the *EMA*) (8, 17, 18).

Currently available prophylactic HPV vaccines are designed using recombinant DNA technology in different expressions systems - insect cells or *Saccharomyces cerevisiae* – and contain L1 Virus Like Particles (VLPs) of certain HPV genotypes (Table II). These expression systems are used for producing L1 antigens of a specific HPV genotype, as the L1 protein has the ability to spontaneously form the so-called VLP that is highly immunogenic and produces high titers of neutralizing antibodies (8). Five L1 monomers self-assemble to pentamers and form capsomeres, and 72 capsomeres further assemble to VLP (Figure 1, 21). Additionally, VLPs are similar to the original HPV, but do not contain viral DNA nor L2 protein, meaning that VLPs are non-infectious and do not have oncogenic potential. HPV prophylactic vaccines containing L1 VLP of certain HPV genotypes are safe to use and there is no possibility that HPV vaccination may cause HPV infection or malignant transformation of human cells.

All approved HPV vaccines protect against highly oncogenic genotypes HPV 16 and HPV 18, which are the most commonly found ones in cervical cancers (19). A reduction in these genotypes, and also in the incidence of genital warts and precancerous cervical lesions, has been achieved in countries with high vaccination coverage. In addition, these results are also detected in non-vaccinated females and males within

countries with high HPV vaccination rates, indicating that herd immunity can also protect non-vaccinated individuals (22).

Table II Types of prophylactic HPV vaccines and vaccine composition of a 0.5 ml dose of HPV vaccine (adapted from 19, 20)

Tabela II Tipovi profilaktičkih HPV vakcina i sastav HPV vakcine u dozi od 0,5 ml (prilagođeno prema referencama 19, 20)

	Gardasil®4 (Merck & Co, Inc., Rahway, NJ, USA)	**Cervarix™** (GlaxoSmithKline Biologicals)	**Gardasil®9** (Merck & Co, Inc., Rahway, NJ, USA)
Year of FDA approval	2006	2009	2014
Year of EMA approval	2006	2007	2015
Valency	4-Valent	2-Valent	9-Valent
Oncogenic protein subunit component L1 VLP (μg)	HPV 6 (20) HPV 11 (40) HPV 16 (40) HPV 18 (20)	HPV 16 (20) HPV 18 (20)	HPV 6 (30) HPV 11 (40) HPV 16 (60) HPV 18 (40) HPV 31 (20) HPV 33 (20) HPV 45 (20) HPV 52 (20) HPV 58 (20)
Adjuvant	0.225 mg aluminum hydroxyphosphate sulfate	ASO4 (0.5 mg aluminum hydroxide and 50 μg 3-O-desacyl-4″-monophosphoryl lipid A (MPL))	0.5 mg aluminum hydroxyphosphate sulfate
Sodium chloride (mg)	9.56	4.4	9.56
L-Histidine (mg)	0.78	/	0.78
Polisorbate 80 (μg)	50	/	50
Sodium borate (μg)	35	/	35
Sodium dihydrogen phosphate dihydrate (mg)	/	0.624	/
Expression system	*Saccharomyces cerevisiae* (yeast)	Baculovirus-insect cell	*Saccharomyces cerevisiae* (yeast)

Abbreviations: FDA (Food and drug administration); EMA (European Medicines Agency); VLP (Virus Like Particle); AS04 (Adjuvant System 04).

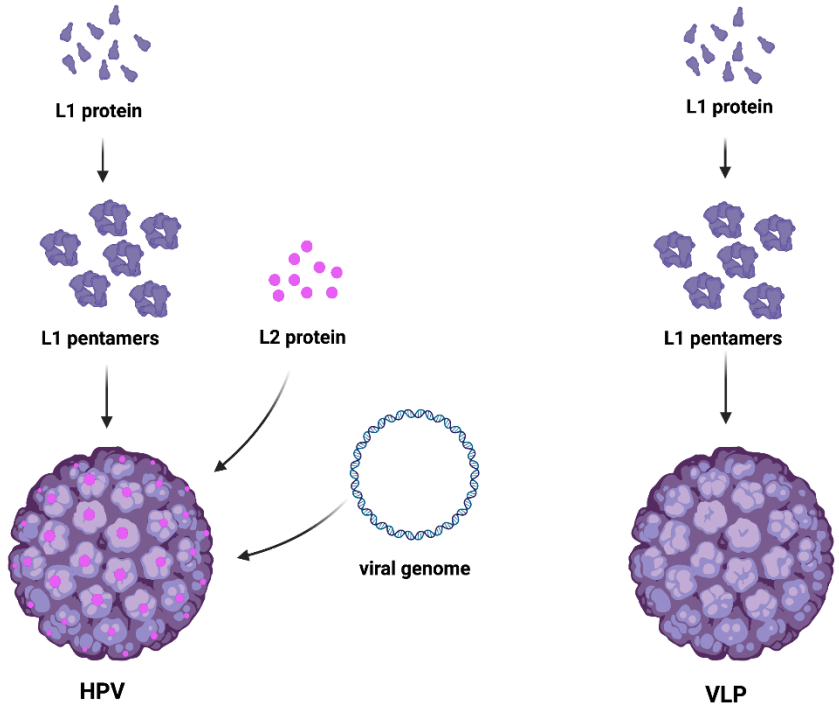

Figure 1. **Comparison of HPV and L1 VLP assembling (created with BioRender.com, adapted from the reference 19)**
Slika 1. **Formiranje HPV i L1 VLP (nacrtano upotrebom BioRender.com programa, prilagođeno iz reference 19)**

Australia was one of the leading countries that were among the first to implement the HPV National Immunisation Programme (NIP) using the quadrivalent HPV vaccine since 2007 for girls, and in 2013 the immunisation programme was extended to boys. This gender-neutral approach is considered vital for disease burden in men, particularly MSM (men who have sex with men) who would not benefit from female-only HPV vaccination. In January 2018, nonavalent HPV vaccine Gardasil®9 replaced the quadrivalent HPV vaccine in Australia (23). The successfully implemented HPV immunisation program in Australia revealed that HPV prevalence in women aged 18-24 declined from 28.7% (before the vaccination programme, from 2005 to 2007) to 2.3% in vaccinated women (from 2010 to 2012) (24). Besides lower HPV prevalence after vaccine introduction, Australia has been one of the leading countries globally in reducing genital warts incidence (25). In addition, a 12 year-long study in Nordic countries (Denmark, Iceland, Norway, and Sweden), where quadrivalent HPV vaccine was introduced, revealed that there were no breakthrough cases of HPV16/18 CIN2 or worse (CIN2+) in 2084 vaccinated women (26).

The nonavalent HPV vaccine is gradually replacing other HPV vaccines in most countries. The highest efficacy of HPV vaccines is among the young population, with the recommendation for vaccination before they become sexually active and before having

contact with HPV. The nonavalent HPV vaccine can be given starting from year 9, but it is best to start with vaccination in children 11-12 years old, and they should get two doses of the HPV vaccine, given 6 to 12 months apart. Children after their 15th birthday need three doses scheduled as 0, 2, and 6 months. The vaccine is indicated according to the EU Summary of product characteristics (SMPC) and local Product Information leaflets for beneficiaries of 9 years of age and above. Routine vaccinations are available according to local jurisdictions through the NIPs or Reimbursement Framework Programs (RFPs). Some countries in the European region, such as Romania and other countries in the EU, provide subsidised programs for adult women from 19 to 45 years of age. In addition, adults between 26 and 45 may consult their physicians regarding the potential benefits of HPV vaccination.

In Serbia, HPV vaccination with nonavalent Gardasil®9 vaccine has been funded by the Republic Fund of Health Insurance for beneficiaries 9-19 years old since June 2022. According to the available data, by the end of 2022, a total of 20,130 doses of Gardasil®9 were administered. The first dose was given to 14,164 people (7,376 to girls and boys 9-14 years old, and 6,788 to people aged 15-19). The second dose was given to 734 people 9-14 years old, while 4,561 second and 618 third doses were given to people aged 15-19 (27).

The vaccine is given as an injection into a muscle, preferably in the shoulder or the thigh, and it is prepared as a suspension for the injection available in vials or prefilled syringes. The side effects are usually mild or moderate and refer to local reactions at the injection site (redness, pain and swelling), and less often fever and headache or fatigue, nausea, muscle pain and syncope (fainting). Although rare, syncope may happen after HPV vaccination, and it is recommended that healthcare professionals administer HPV vaccine while the beneficiary is seated or lying down, and that they wait and observe the beneficiary for 15 minutes to rule out the risk of syncope and possible injury due to a fall. HPV vaccination is contraindicated in pregnant women, people who have had a life-threatening allergic reaction to any component of the HPV vaccine, to a previous dose of the HPV vaccine or yeasts. People with mild infections such as colds may be vaccinated, while vaccination should be postponed in people who are severely ill until they recover (28, 29, 30).

How HPV vaccines work

The immune response induced by natural HPV infection is weak, with very low antibody titres. However, HPV L1 VLPs based prophylactic vaccines have shown great efficacy and effectiveness in clinical trials in countries with high coverage rate. The collected data revealed that inhibitory antibodies (mostly IgG) are the major mediators of vaccine-induced protection. In addition, it is assumed that the specific nature of VLPs is largely attributed to the efficient generation of long-lived antigen-specific antibody-producing cells (31). Several studies have shown that VLPs in HPV vaccine formulations induce effective humoral immune response, and HPV vaccination provides 10- to 100-fold higher antibody titers compared to natural infection (7, 32, 33). Young individuals

aged 9-14 have a better response to HPV vaccination compared to individuals aged 15-24, and antibody titers are stable over time (34). More than 15 years of research has shown that HPV vaccination is safe and effective in reducing HPV-related infections, genital warts and precancerous lesions, and that it provides protection and sustained antibody titers for at least 10 years after vaccination (35).

The exceptional immunogenicity of VLPs based HPV vaccines is largely attributed to the structure of the antigen. VLPs are composed of 360 protein subunits that display a repetitive array of epitopes on their surface and that engage B-cell receptors on naïve B-cells, leading to strong activation of memory B-cells and long-lived plasma cells that produce antibodies for many years (36).

HPV L1 VLPs in vaccines are delivered by intramuscular injection, where dendritic cells of the muscle overtake the antigens and migrate with their cargo to the regional lymph node. This process activates an immune cascade that results in T-cell dependent B cell response and generation of high levels of L1-specific serum neutralizing antibodies and immune memory (37). If natural HPV infection does not involve virus penetration into circulation, the question we may ask is how VLPs based vaccination induces antibodies that reach the HPV infection site, especially women`s genital tract. One possible mechanism is transudation (antibody transport through intact epithelia) of systemic IgG antibodies into the cervicovaginal mucus or direct exudation (passive transfer through damaged epithelia) of systematic antibodies at the site of the infection (38). The very slow progression of natural infection, much slower than for other known viruses, provides an exceptional window for vaccine-induced antibodies to disrupt the process of natural infection (39).

World Health Organization global strategy for the elimination of cervical cancer

In November 2020, the World Health Organization (WHO) announced a global strategy to accelerate the elimination of cervical cancer as a public health problem, a very ambitious global plan whose aim is to reduce the incidence of cervical cancer to less than 4 per 100,000 women worldwide. Why is this global strategy needed? First of all, cervical cancer is a preventable disease, and it can also be adequately treated if detected early. More than 85% of women with cervical cancer are young undereducated women who live in non-developed countries (39). In addition, by the end of 2020, less than 25% of low-income countries and less of 30% lower-middle-income countries had introduced HPV vaccination into their NIP, compared to more than 85% of high-income countries which had implemented NIP in their schedule. To achieve this goal of lowering cervical cancer incidence below 4 per 100,000 women, high coverage of HPV vaccination, screening and treatment of precancerous lesions, and adequate treatment of cervical cancer must be reached by 2030 and maintained at this high level for decades (39).

Therefore, this strategy, known as the Cervical Cancer Elimination Initiative (CCEI) (40), and also as the 90-70-90 strategy, outlines clear targets that must be met by 2030:

- primary prevention – HPV vaccination: 90% of girls will have completed their HPV vaccination course by the age of 15 years;
- secondary prevention - screening: 70% of women will have been screened for cervical cancer using a high-performance test by the age of 35 and again by the age of 45;
- tertiary prevention-treatment: 90% of women diagnosed with cervical disease will have received the treatment (9, 41).

HPV vaccination, as primary prevention, is the most effective long-term intervention for reducing the risk of developing cervical cancer. To achieve 90% coverage of HPV vaccination, different strategic actions are needed: a) securing sufficient and affordable HPV vaccines through appropriate market-shaping interventions; b) increasing the coverage of vaccination (e.g., by implementing school immunization programmes), and additionally, providing monitoring systems or registers in order to track and improve vaccine coverage; c) improving communication and social mobilization efforts, as understanding the social, cultural and other barriers that can affect vaccine acceptance is critical.

The main goal of secondary prevention is to reduce the incidence and mortality rate by identifying and treating women with precancerous lesions. Cytology-based screening has been successfully used with additional diagnostic tests (colposcopy and pathology), but cytology-based programmes have been difficult to implement in low- and middle-income countries. Because of this, initiatives to secure affordable and high-quality diagnostic tests will be prioritized (39).

In addition, comprehensive management of cervical cancer and timely referral of women with suspected or confirmed cervical cancer are crucial for saving lives. Efficient, integrated networks of screening and diagnostic laboratory services are needed, and they will lead to improved access and affordability of screening and treatment, especially in low-income countries (39).

A mathematical model demonstrating the benefits of CCEI implementation by 2030 revealed that the median cervical cancer incidence rate will have fallen by 97% by 2120, preventing more than 74 million new cases of cervical cancer, and 62 million cervical cancer deaths by 2120 (42).

To date, HPV vaccine is part of the NIP for girls in 125 countries and for girls and boys in 47 countries, but this is only about a third of the global population targeted by the 90-70-90 strategy. It is also worrying that global HPV vaccine coverage among girls declined from 20% in 2019 to 15% in 2021, which is far from the goal of the CCEI strategy to vaccinate 90% girls by the age of 15.

In addition, the COVID-19 pandemic disrupted access to preventive strategies, so it remains to be seen whether it is possible to meet the targets of the CCEI or additional efforts will be needed (9).

Treatment of the cervical cancer

One of the goals of the WHO CCEI initiative is to ensure that 90% of women diagnosed with cervical cancer have received appropriate treatment. In 2018, the European Society of Gynecological Oncology (ESGO) jointly published evidence-based guidelines for the management of patients with cervical cancer with the European Society for Radiotherapy and Oncology (ESTRO) and the European Society of Pathology (ESP). In addition, thanks to a large body of new evidence addressing the management of cervical cancer, an update of these evidence-based guidelines was published in 2023 (43).

There are different treatment approaches that depend on the stage of cancer and overall patient health. The selection of treatment plan for cervical cancer nowadays depends on careful preoperative evaluation of pathologic characteristics of the tumour and results of imaging (MRI or expert transvaginal ultrasound). The treatment approach may include one or more different procedures, such as surgery, chemotherapy and/or radiotherapy with external beam radiation therapy and brachytherapy, targeted therapy or immunotherapy (44, 45). Combined therapy – chemoradiotherapy – is the standard of care treatment for locally advanced cervical cancer, and may be effective for many patients; however, the mortality rate is still high (45).

Treatment strategy in early-stage cervical cancer has been modified over the last fifteen years. Radical hysterectomy represents a cornerstone in the treatment of stage IB1 to IIA1. When the surgery is to be performed, careful preoperative evaluation of histopathology, especially the grade and lymphovascular space involvement (LVSI), should be taken into account. Further on, parametrial and/or lymph node involvement diagnosed through imaging methods (MRI or expert transvaginal ultrasound) is another step in planning the treatment protocol. If lymph nodes are radiologically involved, surgery should be abandoned, except for surgical lymph node staging in the paraaortic region. When the sentinel lymph node (SLN) or other lymph nodes are positive on surgical staging, further radical surgery should be abandoned.

After complete pathology of the specimen, the presence of poor prognostic factors such as the size of the tumour, LVSI and depth of stromal invasion (intermediate risk patient) may still demand adjuvant chemoradiotherapy like in the high-risk group of patients (positive lymph nodes, parametria or surgical margins) (45).

Locally advanced cervical cancer should be treated with chemoradiotherapy and brachytherapy. Modern brachytherapy is based on MRI planning that is not available in all countries.

Radiation therapy targets the DNA of cancer cells by high-energy-x rays and it is further divided into external radiation therapy and internal radiotherapy (or brachytherapy) (46).

In those cases where this type of radiotherapy is not developed, the tumour can be pre-treated with neoadjuvant chemotherapy (NACT) for downsizing the tumour. It remains a controversial alternative with no benefits regarding the prognosis (47).

Surgical staging can guide radiation therapy toward the paraaortic region in case of discovery of positive nodes. The debulking can improve sterilisation of the region with radiotherapy. Still, the benefits regarding prognosis after staging and debulking are not completely clarified and demand further randomized studies (43).

Although rare at initial diagnosis, metastatic disease develops in 15-60% of patients with cervical cancer, usually within 2 years of primary treatment. Over the past three decades, the median overall survival (OS) of patients with recurrent and metastatic disease has not improved significantly, despite studies with single-agent or combination chemotherapy. The addition of bevacizumab to chemotherapy (Taxol and Cisplatin) was the only significant advance in the treatment of persistent, recurrent, and metastatic cervical cancer. The results of the GOG 240 study, where the addition of bevacizumab increased median survival by 3.7 months, were the first breakthrough (48).

Chemotherapy is based on drugs used to stop the growth of cancer cells by killing the cells or impacting their ability to divide, and this approach is usually combined with radiotherapy, as mentioned above. Meta analyses from 2017 revealed and confirmed the benefits of concurrent chemoradiotherapy over radiotherapy alone (49).

Thanks to a better understanding of molecular aberrations in cervical cancer, new therapeutic modalities – immunotherapy, including check-point inhibitors, antibody-drug conjugates, and therapeutic vaccines – have appeared in recent years (43).

The addition of pembrolizumab to chemotherapy + bevacizumab in the phase III KEYNOTE-826 study provides a statistically significant improvement in the OS and PFS in persistent, recurrent or metastatic cervical cancer. Pembrolizumab (Keytruda®) is an FDA-approved monoclonal antibody directed against programmed cell death protein 1 (PD-1) which is used for therapy of various cancers, including metastatic or recurrent cervical cancer, following chemotherapy. After a follow-up of 39.1 months, investigators showed that the addition of pembrolizumab to chemotherapy with or without bevacizumab continued to demonstrate clinically a significantly prolonged median OS (26.4 vs 16.8 months; HR: 0.63; P <.0001) and median PFS (10.4 vs 8.2 months; HR: 0.61; P <.0001) in the all-comer population. The median OS was improved in PD-L1 subgroups (PD-L1 CPS ≥1: 28.6 vs 16.5 months, HR: 0.60; PD-L1 CPS ≥10: 29.6 vs 17.4 months, HR: 0.58) (50).

The phase III BEATcc trial of the addition of atezolizumab to the standard of care of bevacizumab and platinum-based chemotherapy as first-line treatment for patients with persistent, recurrent, or metastatic cervical cancer showed that, after a median follow-up of approximately 32 months, the addition of atezolizumab to bevacizumab and chemotherapy improved the median PFS compared with the standard-of-care arm (13.7 vs 10.4 months; HR: 0.62; P = .0001). The PFS benefit in favour of the atezolizumab-containing arm was seen across most subgroups analysed, including age, disease status, chemotherapy backbone, previous chemoradiotherapy, and tumour histology (51).

The antibody-drug conjugate, tisotumab-vedotin, was studied in patients with recurrent and metastatic cervical cancer who had not responded to standard treatment.

Tisotumab vedotin (TivdakTM) is an antibody-drug conjugate (ADC) made of a human monoclonal antibody specific for tissue factor (TF-011) expressed on tumor cells chemically linked to a cancer-killing drug (monomethyl auristatin E; MMAE). In the innovaTV 301 study at the interim primary endpoint of OS, tisotumab vedotin was superior to the investigators' choice of chemotherapy (HR: 0.70; P = .0038). Patients who received tisotumab vedotin experienced a 30% reduction in the risk of death, which is remarkable. At the secondary endpoint PFS was significant (HR: 0.67; P = .0001). The ORR was 17.8% vs 5.2% with tisotumab vedotin vs chemotherapy. Of note, 6 patients (2.4%) in the tisotumab vedotin arm achieved a complete response vs none with chemotherapy (52). Tisotumab vedotin is the first and only ADC recommended for treatment of patients with recurrent or metastatic cervical cancer with disease progression on or after chemotherapy (53).

One of the latest drugs for the treatment of different solid tumours, including cervical cancer, is Cadonilimab, a PD-1/CTLA-4 bi-specific antibody approved in China in June 2022 for use in patients who progressed on or after platinum-based therapy (54).

Therapeutic cervical cancer vaccines aim to eradicate HPV-infected cells by stimulating cytotoxic T cells against viral/tumour antigens. HPV E6 and E7 oncoproteins are expressed in HPV-related cancers and are ideal targets for a therapeutic vaccine (55).

Many other studies have been established to try and reach a better response rate for these advanced/metastatic and recurrent cervical cancers, such as a phase I NRG GY017 study evaluating 3 total doses of atezolizumab given with chemoradiotherapy (CRT) to patients with high-risk LACC, a randomized phase II trial of pembrolizumab during CRT or after CRT in patients with high-risk LACC, a phase III KEYNOTE A18 trial of pembrolizumab with concurrent chemoradiotherapy (cCRT) vs cCRT alone in patients with newly diagnosed, high-risk, previously untreated LACC, a phase III INTERLACE trial evaluating induction chemotherapy with weekly paclitaxel and carboplatin for 6 weeks followed by chemoradiotherapy (CRT) vs CRT alone in patients with newly diagnosed FIGO 2008 stage IB1N+, IB2, II, IIIB, IVA squamous, adeno, and adenosquamous LACC (56-59).

Although effective treatment options have been established, cervical cancer related mortality remains high, especially in low- and middle-income countries with limited availability of treatment options. Great efforts have to be made to reach the WHO goal of having 90% of women diagnosed with cervical cancer receive appropriate treatment.

However, the main goal is decreasing the incidence of cervical cancer, which would lead to a reduction in the need for treatment in advanced disease stages in the future.

Conclusion

Cervical cancer is a significant part of the global cancer burden in women and the fourth cause of cancer-related deaths in woman. The WHO has recognized the importance of reducing and eliminating cervical cancer and announced a global strategy for cervical cancer elimination (CCEI) in 2020, which includes HPV vaccination, screening, and

appropriate treatment. HPV vaccination is a forefront primary preventive measure, and it has been confirmed that HPV vaccination significantly prevents genital warts and cervical cancer. In order to achieve WHO targets, great efforts are needed in the field of improving HPV vaccination awareness, as well as in promoting the importance of regular screenings in women and the availability of chemotherapy and radiotherapy, particularly in low- and middle-income countries.

Conflict-of-interest disclosure

The authors report no potential conflicts of interest. IRM is an employee of Merck Sharp & Dohme Romania SRL.

References

1. Bishop B, Dasgupta J, Chen XS. Structure-based engineering of papillomavirus major capsid l1: controlling particle assembly. Virol J. 2007;4:3.
2. Bedell SL, Goldstein LS, Goldstein AR, Goldstein AT. Cervical Cancer Screening: Past, Present, and Future. Sex Med Rev. 2020;8(1):28-37.
3. Seyoum A, Seyoum B, Gure, T, Alemu A, Alemayehu DH, Alemu A, et al. *H*igh rate of non-vaccine targeted high-risk HPV genotypes circulate among women in Eastern Ethiopia. *Sci Rep*. 2024;14:958.
4. Rosalik K, Tarney C, Han J. Human Papilloma Virus Vaccination. Viruses. 2021;13(6):1091.
5. Brianti P, De Flammineis E, Mercuri SR. Review of HPV-related diseases and cancers. New Microbiol. 2017;40(2):80-5.
6. Human papilloma virus and cancer [Internet]. World Health Organization; 2023 Aug 23 [cited 2023 Dec 23]. Available from: https://www.who.int/news-room/fact-sheets/detail/human-papilloma-virus-and-cancer.
7. HPV and cancer [Internet]. National Cancer Institute [cited 2023 Dec 23]. Available from: https://www.cancer.gov/about-cancer/causes-prevention/risk/infectious-agents/hpv-and-cancer.
8. Mo Y, Ma J, Zhang H, Shen J, Chen J, Hong J, et al. Prophylactic and Therapeutic HPV Vaccines: Current Scenario and Perspectives. Front Cell Infect Microbiol. 2022;12:909223.
9. eClinicalMedicine. Global strategy to eliminate cervical cancer as a public health problem: are we on track? EClinicalMedicine. 2023;55:101842.
10. HPV Centre [Internet]. [cited 2023 Dec 23]. Available from: https://hpvcentre.net/statistics/.
11. Global Cancer Observatory [Internet]. [cited 2024 Jan 15]. Available from: https://gco.iarc.fr.
12. Boon SS, Luk HY, Xiao C, Chen Z, Chan PKS. Review of the Standard and Advanced Screening, Staging Systems and Treatment Modalities for Cervical Cancer. Cancers (Basel). 2022;14(12):2913.
13. Workowski KA, Bolan GA; Centers for Disease Control and Prevention. Sexually transmitted diseases treatment guidelines, 2015. MMWR Recomm Rep. 2015;64(RR-03):1-137. Erratum in: MMWR Recomm Rep. 2015;64(33):924.

14. Tasic D, Lazarevic I, Knezevic A, Tasic L, Pikula A, Perisic Z, et al. The impact of environmental and behavioural cofactors on the development of cervical disorders in HR-HPV-infected women in Serbia. Epidemiol Infect. 2018;146(13):1714-1723.
15. Balasubramaniam SD, Balakrishnan V, Oon CE, Kaur G. Key Molecular Events in Cervical Cancer Development. Medicina (Kaunas). 2019;55(7):384.
16. Schiffman M, Solomon D. Clinical practice. Cervical-cancer screening with human papillomavirus and cytologic cotesting. N Engl J Med. 2013;369(24):2324-31.
17. Hanna E, Bachmann G. HPV vaccination with Gardasil: a breakthrough in women's health. Expert Opin Biol Ther. 2006;6(11):1223-7.
18. European Medicines Agency [Internet]. [cited 2023 Dec 23]. Available from: https://www.ema.europa.eu/en/medicines
19. Yousefi Z, Aria H, Ghaedrahmati F, Bakhtiari T, Azizi M, Bastan R, et al. An Update on Human Papilloma Virus Vaccines: History, Types, Protection, and Efficacy. Front Immunol. 2022;12:805695.
20. Harper DM, DeMars LR. HPV vaccines - A review of the first decade. Gynecol Oncol. 2017;146(1):196-204.
21. Schiller JT, Muller M. Next generation prophy- lactic human papillomavirus vaccines. Lancet Oncol. 2015;16(5):e217–e225.
22. Maver PJ, Poljak M. Progress in prophylactic human papillomavirus (HPV) vaccination in 2016: A literature review. Vaccine. 2018;36(36):5416-23.
23. Patel C, Brotherton JM, Pillsbury A, Jayasinghe S, Donovan B, Macartney K, Marshall H. The impact of 10 years of human papillomavirus (HPV) vaccination in Australia: what additional disease burden will a nonavalent vaccine prevent? Euro Surveill. 2018;23(41):1700737.
24. Tabrizi SN, Brotherton JM, Kaldor JM, Skinner SR, Liu B, Bateson D, et al. Assessment of herd immunity and cross-protection after a human papillomavirus vaccination programme in Australia: a repeat cross-sectional study. Lancet Infect Dis. 2014;14(10):958-66.
25. Drolet M, Bénard É, Pérez N, Brisson M; HPV Vaccination Impact Study Group. Population-level impact and herd effects following the introduction of human papillomavirus vaccination programmes: updated systematic review and meta-analysis. Lancet. 2019;394(10197):497-509.
26. Kjaer SK, Nygård M, Dillner J, Brooke Marshall J, Radley D, Li M, et al. A 12-Year Follow-up on the Long-Term Effectiveness of the Quadrivalent Human Papillomavirus Vaccine in 4 Nordic Countries. Clin Infect Dis. 2018;66(3):339-45.
27. Izveštaj o sprovedenoj imunizaciji na teritoriji Republike Srbije [Internet]. Batut; 2023 [cited 2024 Jan 15]. Available from: https://www.batut.org.rs/download/izvestaji/2022izvestajOSprovedenoj Imunizaciji.pdf.
28. HPPV Vaccine [Internet]. Centers for Disease Control and Prevention [cited 2023 Dec 23]. Available from: https://www.cdc.gov/hpv/parents/vaccine-for-hpv.html.
29. Gardasil 9 [Internet]. European Medicines Agency [cited 2023 Dec 23]. Available from: https://www.ema.europa.eu/en/medicines/human/EPAR/gardasil-9.
30. Human Papillomavirus (HPV) Vaccine [Internet]. Centers for Disease Control and Prevention [cited 2023 Dec 23]. Available from: https://www.cdc.gov/vaccinesafety/vaccines/hpv-vaccine.html.

31. Schiller J, Lowy D. Explanations for the high potency of HPV prophylactic vaccines. Vaccine. 2018;36(32 Pt A):4768-73.
32. Pattyn J, Van Keer S, Tjalma W, Matheeussen V, Van Damme P, Vorsters A. Infection and vaccine-induced HPV-specific antibodies in cervicovaginal secretions. A review of the literature. Papillomavirus Res. 2019;8:100185.
33. Villa LL, Costa RL, Petta CA, Andrade RP, Paavonen J, Iversen OE, et al. High sustained efficacy of a prophylactic quadrivalent human papillomavirus types 6/11/16/18 L1 virus-like particle vaccine through 5 years of follow-up. Br J Cancer. 2006;95(11):1459-66.
34. Romanowski B, Schwarz TF, Ferguson L, Peters K, Dionne M, Behre U, et al. Sustained immunogenicity of the HPV-16/18 AS04-adjuvanted vaccine administered as a two-dose schedule in adolescent girls: Five-year clinical data and modeling predictions from a randomized study. Hum Vaccin Immunother. 2016;12(1):20-9.
35. Shapiro GK. HPV Vaccination: An Underused Strategy for the Prevention of Cancer. Curr Oncol. 2022;29(5):3780-92.
36. Schiller JT. The Biological Rationale for a One Dose HPV Vaccine [Internet]. HPW: The newsletter on HPV [cited 2023 Dec 23]. Available from: https://www.hpvworld.com/articles/the-biological-rationale-for-a-one-dose-hpv-vaccine/.
37. Stanley MA, Sterling JC. Host responses to infection with human papillomavirus. Curr Probl Dermatol. 2014;45:58-74.
38. Schiller JT, Lowy DR. Understanding and learning from the success of prophylactic human papillomavirus vaccines. Nat Rev Microbiol. 2012;10(10):681-92.
39. Global strategy to accelerate the elimination of cervical cancer as a public health problem [Internet]. World Health Organization [cited 2024 Jan 15]. Available from: https://iris.who.int/bitstream/handle/10665/336583/9789240014107-eng.pdf?sequence=1.
40. Cervical Cancer Elimination Initiative [Internet]. [cited 2023 Dec 23]. Available from: https://cceirepository.who.int.
41. Cervical Cancer Elimination Initiative [Internet]. World Health Organization [cited 2023 Dec 23]. Available from: https://www.who.int/initiatives/cervical-cancer-elimination-initiative.
42. Global strategy to accelerate the elimination of cervical cancer as a public health problem [Internet]. World Health Organization; 2020 Nov 17 [cited 2023 Dec 23]. Available from: https://www.who.int/publications/i/item/9789240014107.
43. Cibula D, Raspollini MR, Planchamp F, Centeno C, Chargari C, Felix A, et al. The European Society of Gynaecological Oncology/European Society for Radiotherapy and Oncology/European Society of Pathology Guidelines for the Management of Patients With Cervical Cancer. Int J Gynecol Cancer. 2023;33:649–66.
44. PDQ Cancer Information Summaries [Internet]. Bethesda (MD): National Cancer Institute (US); 2002-. Cervical Cancer Treatment: Patient Version; 2023 Nov 2 [cited 2023 Dec 23]. Available from: https://www.ncbi.nlm.nih.gov/books/NBK65985/
45. Mayadev JS, Ke G, Mahantshetty U, Pereira MD, Tarnawski R, Toita T. Global challenges of radiotherapy for the treatment of locally advanced cervical cancer. Int J Gynecol Cancer. 2022;32(3):436-45.

46. Cervical Cancer Treatment [Internet]. National Cancer Institute [cited 2023 Dec 23]. Available from: https://www.cancer.gov/types/cervical/treatment.
47. Yang SL, Chen L, He Y, Zhao H, Wu YM. Effect of neoadjuvant chemotherapy followed by surgery for FIGO stage I-II cervical cancer: a meta-analysis. J Int Med Res. 2020 Aug;48(8):300060520945507.
48. NCCN [Internet]. Clinical practice guidelines in oncology: cervical cancer. v.1.2022 [cited 2023 Dec 23]. Available from: nccn.org.
49. Shrivastava S, Mahantshetty U, Engineer R, Chopra S, Hawaldar R, Hande V, et al. Cisplatin Chemoradiotherapy vs Radiotherapy in FIGO Stage IIIB Squamous Cell Carcinoma of the Uterine Cervix: A Randomized Clinical Trial. JAMA Oncol. 2018;4(4):506-13.
50. Monk BJ, Colombo N, Tewari KS, Dubot C, Caceres MV, Hasegawa K, et al. KEYNOTE-826: Final overall survival results from a randomized, double-blind, phase 3 study of pembrolizumab + chemotherapy vs placebo + chemotherapy for first-line treatment of persistent, recurrent, or metastatic cervical cancer. J Clin Oncol. 2023;41:5505-5511.
51. Oaknin A, Gladieff L, Martinez-Garcia J, Villacampa G, Takekuma M, De Giorgi UFF, et al. Primary results from BEATcc (ENGOT-Cx10/ GEICO 68-C/JGOG1084/GOG-3030), a randomised phase III trial of first-line atezolizumab (atezo) combined with a platinum doublet and bevacizumab (bev) for metastatic (stage IVB), persistent or recurrent cervical cancer. ESMO virtual plenary abstracts. 2023;34(12):P1198-1200.
52. Vergote I, González-Martín A, Fujiwara K, Kalbacher E, Bagameri A, Ghamande S, et al. InnovaTV 301/ENGOT-cx12/GOG-3057: a global, randomized, open-label, phase 3 study of tisotumab vedotin vs investigator's choice of chemotherapy in 2L or 3L recurrent or metastatic cervical cancer. Ann Oncol. 2023;34:S1276-7.
53. Markham A. Tisotumab Vedotin: First Approval. Drugs. 2021;81(18):2141-47.
54. Keam SJ. Cadonilimab: First Approval. Drugs. 2022;82(12):1333-9.
55. Youn JW, Hur SY, Woo JW, Kim YM, Lim MC, Park SY, et al. Pembrolizumab plus GX-188E therapeutic DNA vaccine in patients with HPV-16-positive or HPV-18-positive advanced cervical cancer: interim results of a single-arm, phase 2 trial. Lancet Oncol. 2020;21(12):1653-60.
56. Zamarin D, Deng W, Lankes H, Ghamande S, Holman L, Mathews C, et al. Evolution of peripheral and tumor immune parameters as biomarkers of differential sequencing of immune checkpoint blockade and chemoradiotherapy in locally advanced cervical cancer: an NRG Oncology/GOG study. Gynecol Oncol. 2023;176(1S):S6.
57. Duska LR, Showalter TN, Petroni GR, Bullock T. A randomized phase II study of chemoradiation and pembrolizumab for locally advanced cervical cancer. J Clin Oncol. 2017;35:TPS5601-TPS5601.
58. Lorusso D, Xian Y, Hasegawa K, Scambia G, Leiva Galves MH, Ramos Elias P, et al. Pembrolizumab plus chemoradiotherapy for high-risk locally advanced cervical cancer: the randomized, double-blind, phase 3 ENGOT-cx11/GOG-3047/KEYNOTE-A18. Ann Oncol. 2023;34:S1279-80.
59. McCormack M, Gallardo Rincón D, Eminowicz G, Diez P, Farrelly L, Kent C, et al. A randomised phase III trial of induction chemotherapy followed by chemoradiation compared with chemoradiation alone in locally advanced cervical cancer. The GCIG INTERLACE trial. Ann Oncol. 2023;34:S1276.

Humani papilomavirusi i karcinom grlića materice iz perspektive inicijative Svetske zdravstvene organizacije za eliminaciju karcinoma grlića materice

Brankica Filipić[1]*, Ivana Rapajić-Moran[2], Ines Nikolić[3,4], Slavica Oljačić[5], Aljoša Mandić[6,7]

[1]Katedra za mikrobiologiju i imunologiju, Univerzitet u Beogradu – Farmaceutski fakultet, Vojvode Stepe 450, 11221 Beograd, Srbija

[2]Katedra za socijalnu farmaciju i farmaceutsko zakonodavstvo, doktorske studije, Univerzitet u Beogradu – Farmaceutski fakultet, Vojvode Stepe 450, 11221 Beograd, Srbija

[3]Katedra za farmaceutsku tehnologiju i kozmetologiju, Univerzitet u Beogradu – Farmaceutski fakultet, Vojvode Stepe 450, 11221 Beograd, Srbija

[4]School of Pharmaceutical Sciences, University of Geneva – Faculty of Science

[5]Katedra za farmaceutsku hemiju, Univerzitet u Beogradu – Farmaceutski fakultet, Vojvode Stepe 450, 11221 Beograd, Srbija

[6]Odeljenje ginekološke onkologije, Institut za onkologiju Vojvodine, Put dr Goldmana 4, 21204 Sremska Kamenica, Srbija

[7]Katedra za ginekologiju i akušerstvo, Univerzitet u Novom Sadu – Medicinski fakultet, Hajduk Veljkova 3, 21137 Novi Sad, Srbija

*Autor za korespondenciju: Brankica Filipić, e-mail: brankica.filipic@pharmacy.bg.ac.rs

---

Kratak sadržaj

Humani papilomavirusi (HPV) su među najčešćim uzročnicima seksualno prenosivih patogena i mogu dovesti do različitih kliničkih manifestacija: od benignih stanja do različitih vrsta karcinoma kod žena, ali i muškaraca. Najčešći HPV-posredovan karcinom je karcinom grlića materice koji je u preko 99,99% slučajeva posledica infekcije. Najefikasniji način da se spreči razvoj perzistentne HR-HPV infekcije je vakcinacija. Dostupne su tri profilaktičke vakcine: dvovalentna, kvadrivalentna i devetovalentna. Devetovalentna vakcina pruža najširu zaštitu jer sadrži devet onkogenih HPV genotipova i postepeno zamenjuje ostale vakcine u svim zemljama. Sa vakcinacijom se može krenuti od 9. godine, ali se najčešće rutinski sprovodi kod dečaka i devojčica u uzrastu od 11 do 12 godina. Svetska zdravstvena organizacija je prepoznala karcinom grlića materice kao globalni problem i uvela takozvanu 90-70-90 strategiju u cilju smanjenja stope, pa čak i eliminacije karcinoma grlića materice. Ova strategija podrazumeva da 90% devojčica bude potpuno vakcinisano do 15. godine, 70% žena pristupi redovnom ginekološkom

pregledu do 35. godine i ponovo do 45. godine i 90% žena sa promenama na grliću materice primi adekvatnu terapiju. Iako su dostupne različite terapije poput hirurškog tretmana, radioterapije, hemioterapije i ciljane terapije monoklonskim antitelima, i dalje su potrebni veliki napori da bi se dostigli ciljevi Svetske zdravstvene organizacije.

Ključne reči: HR-HPV infekcija, karcinom grlića materice, HPV vakcinacija

---