Cervical Cancer and Genital Infections: Assessment of Performance and Validation in Human Papillomavirus Genotyping Assays in Iran, its Neighbouring Countries and Persian Gulf Area

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**KEYWORDS**

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**ABSTRACT**

**Background:** The accuracy of diagnostic assays in Human Papillomavirus (HPV) genital infection and cervical cancer has remained a clinical challenge in diagnosis. Evidence indicates that a large proportion of cervical cancer can be prevented through organized care for HPV and testing. Countries with low per capita income, such as Iran and its neighbours, have no national organized program for cervical cancer screening and vaccination. The aim of this study was to review recent published papers in this region for evaluating the efficacy of released data regarding HPV genotyping system in genital infections and cervical cancer

**Methods:** Investigating various medical search engines retrieved 46 reports, mostly after 2010, consisting of either home brew protocols or commercial technologies in this field.

**Results:** Summarized results demonstrated that except a few cases, all reports were limited studies performed in confined populations focusing on attending patients at clinics for regular checkups. In the present study, 52.8% of papers were from Iran and the rest belonged to other countries. The rate of HPV infection was reported in the range of 0.62% to 25% in the normal population, while it varied from 18.75% to 100% in females with cervical cancer. In HPV genotyping surveys, only 26.1% (12/46) of reports had validated and World Health Organization (WHO) proficient procedures. Also, multiple infections were not mentioned in 56.52% (25/46) of researches.

**Conclusions:** Employing reliable genotyping methods is the best way for regular screening of cervical cancer related to HPV and precancerous diseases in females of these areas. The focus of most surveys was to come up with the best national policies for establishing a preventive program in Iran and Persian Gulf area.

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Introduction

More than 200 Human Papillomavirus (HPV) genotypes have been detected, approximately 40 of which can infect the genital tract. Infection with high-risk HPV is considered a major cause of cervical cancer (1,2). Based on WHO guidelines, approximately sixteen HPV genotypes have been diagnosed in laboratories (HPVs 6,11 [Low Risks], 16,18,31, 33,35,39,45,51,52,56,58,59,66,68 [High Risks]). Human Papillomavirus genotyping test refers to HPV IVD devices, which detect and should be able to differentiate between high-risk and low-risk HPV genotypes (3, 4). Al-Awadhi et al. 2011, underlined that most females with normal cytology could be infected with low and high-risk genotypes (5). Several commercial technologies are available such as Digene Hybrid Capture 2 High-Risk HPV DNA...
Test, Cervista™ HPV HR and Genfind™ DNA Extraction Kit, Cervista™ HPV 16/18, Cobas HPV Test and APTIMA® HPV Assay, which are Food and Drug Administration (FDA) approved (4,6). However, improving the efficacy of the diagnostic methods for HPV detection and genotyping assays is still one of the main objectives in this field. It seems that DNA-based methods for HPV detection and genotyping are mostly designed for L1, E6/E7, E1 and E2 of HPV genome. These regions are more sensitive and specific than others. In most diagnostic commercial assays, the L1 region is used. The L1 region has conserved and variable sequences, therefore it is suitable for HPV detection, genotyping and classification (6-9). Abru et al. have reviewed the current HPV genotyping methods. They underlined that despite several methods being available and some molecular techniques being developed, HPV protocols still need to become more rapid, automated, and cost effective, to be of practical use in low income countries (7). Besides an assessment of the regional distribution of HPV genotypes and accurate evaluation of human papillomavirus infections, genotyping is still of critical importance to evaluate the epidemiology of HPV infections worldwide and to establish and monitor HPV vaccine efficacy. Reporting true negative results has an important role in decreasing the prevalence of the HPV. It is important to note that negative results of HPV DNA may not negate HPV infection, and failure of HPV diagnostic methods and incorrectly interpreted results may lead to incorrect treatment and cervical cancer related to HPV screening (10).

The cost of these assays effects the selection of the HPV genotyping methods in both developing and under developed countries.

Discrimination between the high and low risk genotypes of HPV in clinical diagnostic exposures is one of the concerns in developing countries, and countries that do not have access to new technologies (7-9). This study reviewed researches related to Iran's neighbouring countries and the Persian Gulf area, which had focused on HPV genotyping technologies and prevalence of HPV in single and multiple genital infections and cervical cancer.

Materials and Methods

Study Design

This Meta-analysis focused on recent HPV reports (2010 to 2015) in our region. This review included all researches on genital infections and cervical cancer related to HPV from Iran, Pakistan, Afghanistan, Iraq, Kuwait, Saudi Arabia, Bahrain, Qatar, Oman, Yemen, Jordan and Syria. Northern neighbouring countries of Iran were excluded from this study, as they belong to Russian Federation countries (the western countries of the former Soviet Union, Caucasus Region and central Asia).

Data Sources

We searched several relevant international medical databases including PubMed, Scopus, and Google Scholar, as well as local databases. We also checked the reference lists of retrieved articles to identify additional studies.

Data Extraction

The data were extracted and summarized. The final eligible articles were reviewed by both authors and disagreements were resolved by consensus. The following study characteristics were extracted, first author, year of publication, region of the study, studied group, detection method, and analyzed results. The applied method was assessed by accepted international approved guidelines and protocols such as WHO and Food and Drug Administration (FDA). Papers with no accurate method or results were excluded from the review. Necessary items for qualified technique were as follows:

1. Sample Preparation Methods

Application of a proper method for specimen collection and shipment had critical roles in HPV results. Cellular samples from the site of infection and cervical transformation zone are the best specimens for molecular diagnosis in both cervical cancer and genital infections. We should be aware that suitable specimens are those with infected cells such as Liquid Based Cytology specimens (LBCs), Formalin Fixed Paraffin Embedded Tissue (FFPE), or a portion of genital warts and lesions. Those specimens stored at room temperature (~ ≥14 days) or at 4°C for ~ ≥three weeks are not recommended for testing. The desired storage condition is -20°C for long durations (3,4).
2. Home Brew Human Papilloma Protocols

The clinical performance of a qualitative test (test with two outcomes, positive or negative) should be described by its clinical sensitivity and specificity. The results of home brew protocols are required to be established in prospective clinical studies, since the results are going to be considered as final decisions for HPV infections. All effective criteria for true results should be evaluated and determined according to the FDA and WHO guidelines. At present, PGMY, GP5/GP6, MY09/MY11 and SPF10 general primers are used in most commercial and home-brew diagnostic systems, which are validated and utilized by WHO and companies as screening methods. These primer pairs amplify part of the L1 HPV genome as L1 consensus Polymerase Chain Reaction (PCR). The rest of primers, which are used in research projects and clinical laboratories, should be qualified and participated in proficiency study and quality assessment programs.

3. Differentiation of True Negative from False Negative

Reporting false negative and positive results could lead to missing proper treatment strategies and increasing morbidity and mortality of cervical cancer. Applying an Internal Control (IC) is considered to be mandatory for diagnosis. False positive results may be due to contamination, carry over and sampling cross contamination. False negative results could be affected by low virus copy number, sampling and specimen collection errors. The use of Oral Contraceptive Pills (OCPs), topical ointments, cryotherapy and laser therapy could be regarded as inhibitory for amplification and may interfere with DNA-based methods.

4. Low and High Risk Types

It has been frequently reported that low risk types have a considerable role in genital infections and even cervical cancer with low percentage. These low risk types, especially HPVs 6 and 11, are relatively common in atypical squamous cells of undermined significance (ASCUS), although can play a role in mixed infections. Therefore, detection of dominant genotypes must be included in any diagnostic protocol (11). Fourteen HPV high-risk types (16,18,31,33,35,39,45,51,52,56,58,59,66,68) plus HPV 6 and 11 are included in proficiency panel testing of WHO.

5. The World Health Organization Human Papillomavirus LabNet

The WHO in collaboration with the Karolinska Institute in Sweden has established the HPV LabNet. Evaluation on performance of HPV diagnostic methods, during annual proficiency testing program, is performed, especially in Reference Health Laboratories around the world. Commercial and in-house diagnostic assays for HPV genotyping are evaluated. Results of this program are the best reference for better understanding of HPV genotyping methods performance. Hence, in the present study, based on these results, efficiency, validation and comparison of HPV genotyping technologies are appraised.

Results

Various search engines revealed 46 reports, performed mostly after 2010. The number of applied commercial kits or home brew protocols was different in each area of our region (Table 1 and 2). Valid general primers such as GP5/6 and MY09/MY11 for general HPV detection were used in 41.3% (19/46) of published articles and 58.7% (27/46) used other sequences. It seems that most procedures for HPV genotyping methods were not mentioned in WHO and FDA guidelines in these areas. We considered HC2, LiPA, Roche, and GenoArray commercial diagnostic kits as approved genotyping methods. Other used genotyping methods were unconfirmed protocols or did not fully cover all necessary types such as some PCR hybridization methods, PCR sequencing, multiplex PCR, and Real Time PCR method. Only one home-brew HPV genotyping method, In-House Multiplex Real time PCR, was approved in WHO proficiency testing among these reports (12). Overall, 13 out of 46 papers used protocols covering all necessary detected types consisting of 10 approved methods, 2 commercially unapproved procedures and one home brew developed method (Table 1 and 2). Studied population reports were based on patients attending clinics for regularly checkups, cervical cancer, abnormal cytology, negative pap smears, genital wart, and HPV positive samples were found in 16, 14, 3, 1, 1 and 1 reports, respectively. Ten papers had established their investigation on a mixed source of specimens. The prevalence of HPV in patients, who attended for routine checkups varied from 0.62% to 25% in performed researches. Those reports on
specimens collected from patients with cervical cancer specimens also had variable prevalence from as low as 18.75% to 90.76% and even 100%. Multiple infections were recorded in some articles; 20 reports (63.04%) out of 46 investigates, 10 from Iran and 10 from Arabic countries but none from Pakistan. Our search retrieved no data from Jordan, Oman and Afghanistan in scientific medical databases.

### Table 1: Human Papillomavirus Genotypes Distribution in Genital Infection and Cervical Cancer in Iran

| Author/Year          | Region                  | HPV Detection Method                  | Number / Type of Study                                      | HPV Positive / HPV Genotypes / Co-infection |
|----------------------|-------------------------|---------------------------------------|------------------------------------------------------------|---------------------------------------------|
| Hamzavi et al., 2002 | Mazandaran/ Iran         | Home-brew PCR                         | 100 / Routine Checkups & Cervical Cancer                   | 78.6% / HPV 16,18,31,35,6,31,11 / HPV 16/18: 1.51%, HPV31/33: 16.24%, HPV6/11: 4.18% |
| Mortazavi et al., 2002 | Tehran/Im               | In Situ Hybridization & Home Brew PCR (Triplex PCR 16,18,33) | 100 / FFPE Cervical Cancer                                  | 82% / HPV 16,18,3 / Not Mentioned           |
| Hamzavi et al., 2003 | Mazandaran/ Iran         | Home-brew PCR                         | 254 / Routine Checkups & Cervical Cancer                   | 44.48% / HPV 16,6,11,31,33 / Not Mentioned  |
| Farjadani et al., 2003 | Shiraz/ Iran             | Home-brew PCR                          | 101 / FFPE Cervical Cancer                                  | 87.12% / HPV16,18 / Not Mentioned           |
| Zandi et al., 2010   | Bushehr/ Iran            | Home Brew PCR GP5/GP6 & Sequencing     | 200 / Routine Checkups                                      | 5.5% / HPV 16,18,53 / Not Mentioned         |
| Safaei et al., 2010  | Shiraz/ Iran             | Home-brew PCR (Duplex PCR 16k18)      | 402 / Routine Checkups                                      | 5.47% / HPV 16 / Not Mentioned              |
| Shahramian et al., 2011 | Zabol/ Iran             | Home brew PCR GP5/GP6 - MY09/MY11 & Duplex PCR 16k18 | 402 / Routine Checkups                                      | 1.2% / HPV 16,18 / Not Mentioned           |
| Jafari et al., 2011  | Shiraz/ Iran             | Real-time for 16,18,33,52 (Primer Design®) | 100 / Tissue, Biopsy Blood & Genital Wart                  | 19% / HPV 16,18 / 2%                        |
| Jafari et al., 2011  | Shiraz/ Iran             | Real-time for 16,18,33,52 (Primer Design®) | 79 / Genital Wart                                           | 16.45% / HPV 16,18 / 1.3%                   |
| Moradi et al., 2011  | Gorgan/ Iran             | Home Brew PCR GP5/GP6 - MY09/MY11 & Duplex PCR 16k18 | 308 / Routine Checkups                                      | 24.67% / HPV 16,18 / Not Mentioned         |
| Allameh et al., 2011 | Isfahan/ Iran            | Home Brew PCR GP5/GP6 & Primer Specific PCR HPV 6,11,16,18 | 130 / Abnormal Cytology                                    | 24.67% / HPV 16,18,6,11 / 83.1 %           |
| Shamsia et al., 2011 | Tehran/ Iran             | Home Brew PCR GP5/GP6 & Sequencing     | 87 / FFPE Cervical Cancer                                   | 79.31% / HPV 16,18,31,45 / Not Mentioned   |
| Khodakarami et al., 2011 | Tehran/ Iran           | Home Brew PCR GP5/GP6                  | 825 / Normal & Abnormal Cytology                             | 7.75% / HPV 16,18,31,39,45,51,52,56,58,59,73,63,40,42,54,66,67 / ≥3% |
| Tghibli et al., 2012 | Bushehr/ Iran            | Home Brew PCR GP5/GP6 & LiPA           | 790 / Routine Checkups                                      | 0.62% / HPV 16,18,31,33,51,56,66 / 0.25%   |
| Hamidifard et al., 2012 | Alborz/ Iran            | Home Brew PCR GP5/GP6 & Sequencing     | 60 / FFPE (Normal & Abnormal)                               | 13.33% / HPV 16,18,6,11 / Not Mentioned   |
| Meeni et al., 2012   | Tehran/ Iran             | Home Brew PCR MY09/MY11               | 96 / FFPE Cervical Cancer                                   | 18.75% / HPV Positive / Not Mentioned      |
| Haghshenas et al., 2013 | Sari/ Iran              | Amplisens®                             | 98 / FFPE Cervical Cancer                                   | 79.59% / HPV 16,18,45,39 / Not Mentioned   |
| Shafaghi et al., 2013 | Tehran/ Iran             | Home Brew Nested PCR MY09/MY11, GP5/GP6 & RFLP | 851 / Routine Checkups                                      | 30.08% / HPV16,18,31,33,35,39,45,51,52,56,58,68,66,68,61,42,43,44,29,1% |
| Mobinnekhesa et al., 2013 | 11 Province/ Iran      | Home Brew Nested PCR Only HPV 16       | 108 / Routine Checkups                                      | 25% / HPV16 / Not Mentioned                |
| Shameezaazadeh et al., 2014 | Isfahan/ Iran           | Home Brew PCR GP5/GP6 & Duplex PCR 16k18 | 150 / FFPE Cervical Cancer                                  | 58.97% / HPV 16,18 / 3.85%                 |
| Pouryasin et al., 2014 | Tehran/ Iran            | LiPA                                   | 80 / HPV Positive Patients                                  | 100% / 6,16,53,18,52,39,66,11,31,33,35,45,56,58,68,82,54,44,69/1 / Co-infection 50% |
| Mirzaei-Kashani et al., 2014 | Isfahan/ Iran           | Home Brew Nested PCR MY09/MY11, GP5/GP6 | 122 / FFPE Cervical Cancer                                  | 76.39% / HPV Positive / Not Mentioned      |
| Sehrabi et al., 2014 | Tehran/ Iran             | Home Brew TaqMan Real Time PCR         | 112 / Routine Checkups & Cervical Cancer                    | 93.75% / HPV 6,11,16,18,31,33,35,39,45,51,52,56,58,59,68 / ≥124.9% |
| Mohseni-Mehran et al., 2015 | Rasht/ Iran             | Home Brew PCR GP5/GP6                 | 103 / Routine Checkups & Abnormal Cytology                  | 4.9% / HPV Positive / Not Mentioned        |
Table 2. Human Papillomavirus Genotypes Distribution in Genital Infection and Cervical Cancer in Iran’s Neighbouring Countries and Persian Gulf Area

| Author/Year | Region       | HPV Detection Method                                      | Number / Type of Study | HPV Positive/ HPV Genotypes / Co-infection |
|-------------|--------------|----------------------------------------------------------|------------------------|------------------------------------------|
| Raza et al., 2010 [36] | Pakistan     | Home Brew PCR GP5/GP6 & Reverse Line Blot Hybridization | 899/ Routine Checkups & Abnormal Cytology | 2.78% / HPV 16,18,45,56,33,59 / Not Mentioned |
| Siddique et al., 2014 [37] | Pakistan     | Home Brew PCR GP5/GP6 & Duplex PCR 16&18              | 77/ Cervical Cancer | 53.24% / HPV16,18 / Not Mentioned |
| Akbar et al., 2015 [38] | Pakistan     | Home Brew PCR GP5/GP6 & Duplex PCR 16&18              | 200/ Routine Checkups | 2% / HPV 16,18 / Not Mentioned |
| Shahid et al., 2015 [39] | Pakistan     | Home Brew PCR GP5/GP6 & Duplex PCR 16&18              | 160/ Routine Checkups | 1.87% / HPV 16 / Not Mentioned |
| Darnel et al., 2010 [40] | Syria        | Home Brew Specific PCR (E7)                            | 44/ FFPE Cervical Cancer | 95.45% / HPV 33,16,18,45,52,58,35,51,31 / Not Mentioned |
| Banna et al., 2014 [41] | UAE          | Home Brew& mix of 13 primers                           | 50/ Neg. Pap Smear Tests | 10% / Any of 13 types / Not Mentioned |
| Bensumande et al., 2014 [42] | Yemen        | Immunohistochemistry & HPV16/18 kit (Sacace-Biotechnologies) | 84/ Cervical Cancer | 100% / HPV 16,18 / Not Mentioned |
| Ahmed et al., 2015 [43] | Yemen        | HPV 31,33,35,39,45 (Sacace-Biotechnologies)            | 200/ FFPE Cervical Cancer & Benign Tissue | 24%/HPV 31,45,33,35,39/ 1% |
| Haniya et al., 2006 [44] | Bahrain      | Home Brew PCR & RFLP                                      | 100/ Routine Checkups | 11% / HPV 16,18,45,62,53 / Not mentioned |
| Moosa et al., 2014 [45] | Bahrain      | LiPA                                                     | 571/ Routine Checkups | 9.8% / HPV52,16,31,51,6,70,74 / 2.62% |
| Fadhl et al., 2014 [46] | Iraq         | Genoaarray primer-based PCR 21 HR & LR types          | 188/ Cervical Dysplasia | 99.91% (82.35% HR-types & 17.56% LR types) / 27.94% |
| Al-Awadih et al., 2011 [47] | Kuwait       | Home Brew PCR GP5/GP6, MY09/MY11 & Sequencing         | 301/ Routine Checkups | 2.3%/HPV11,81,61,53,54,90,31,73,84,106,33,56,58,67,70,82,83,10 / Not Mentioned |
| Al-Tahani et al., 2010 [48] | Qatar        | RT-PCR (E1–E2 region) Sacace Biotechnologies, Italy   | 95% / Abnormal Screening | 64% / HPV 16,52,56 / 34.4% |
| Elm et al., 2012 [49] | Qatar        | Real Time PCR Sacace Biotechnologies, Italy            | 1100/ Routine Checkups | 11.36%/15 HPV Genotypes Detected / Not Mentioned |
| Bansal et al., 2014 [50] | Qatar        | Real Time PCR Sacace Biotechnologies, Italy            | 3008/ Routine Checkups | 11.8% / HPV 81,11,16,18,56 / 0.93% |
| Al-Sheib et al., 2011 [51] | Saudi Arabia | Linear Array Detection Kit (LA DK) (Roche) (37 genotypes) | 100/ Cervical Cancer | 89% / HPV 16,18,31,39,45,51,59,73,6,64,7 0 / 5.5% |
| AlBadawi et al., 2011 [52] | Saudi Arabia | Home Brew PCR GP5/GP6,MY09/MY11 & Sequencing & Reverse Line Blot Hybr (RLB) | 90/ Cervical Cancer | 95.55%/HPV 16,18,45,33,31,52,58,59,66 /18.88% |
| Al-Sheib et al., 2013 [53] | Saudi Arabia | Linear Array HPV Genotyping (Roche) (37 genotypes)     | 318/ Cervical Cancer | 82% / HPV 16,31,18,45,33,32,70,592,59 / 2.2% |
| Bondy et al., 2013 [54] | Saudi Arabia | Hybrid Capture II                                       | 485/ Routine Checkups | 5.6% / Not Mentioned |
| Turki et al., 2013 [55] | Saudi Arabia | Multiplex PCR Plus BiolabChip Hybridization Assay (26 types) | 40/ Abnormal Screening | 42.5% / HPV 16,18,45 / 0% |
| Al-Obaid et al., 2014 [56] | Saudi Arabia | LiPA                                                     | 417/ Routine Checkups | 6.95% / HPV68/31,18,31,51,52,39,5 6.58,6 42,54,11,40,70,74 / 0.95% |
| Al-Ahbal et al., 2014 [57] | Saudi Arabia | Reverse Line Blot Hybridization                         | 519/ Routine Checkups & Abnormal Cytology | 31.6% / HPV16,18,17 / Not Mentioned |

Discussion

WHO/International Information Center on HPV (ICO) and cervical cancer reports that more than 99% of cervical cancers are associated with HPV infection (3). An organized prevention program is frequently reported to be needed for reducing the rate of cervical cancer in low and middle income countries (56). It is frequently reported that considerable number of females diagnosed with cervical cancer have never been screened by HPV testing or they may even be asymptomatic patients (39,58-60). Human Papillomavirus infection is diagnosed mainly by molecular methods and can be either DNA-based or RNA assays. The DNA-based assays are the best method for HPV detection and typing, especially the early stages of infection since

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their sensitivity are relatively higher than RNA based methods. However, once the disease has progressed, RNA-based assays are considered more accurate and recommended for cervical cancer infections. Human Papillomavirus DNA testing has been useful in decreasing mortality rates, which are due to cervical cancer up to 50% in developed countries (61-63). The usefulness, specificity, sensitivity and cost effectiveness of HPV DNA testing and genotyping have been compared to visual inspection methods for cervical cancer screening, which are reported to achieve the maximum effectiveness for patients, who are between 35 to 50 years old (62-64). An appropriate technique for use in clinical laboratory depends on parameters such as the cost of the assay, population to whom this test is to be applied, as well as specificity and sensitivity of the test. However, in under developed countries, cost effectiveness may be the most important factor. Human Papillomavirus genotyping methods merit attention in several aspects. Polymerase Chain Reaction hybridization and conventional amplification-based approaches have been used for HPV screening and genotyping. Each of these methods has some drawbacks, mainly in the area of reagents, cost effectiveness, time required, and false positive and negative results, especially in samples with lower copy numbers and mixed infections. Results of some surveys show that primers such as MY09/11 are ineffective due to increased mutations in HPV genome and multiple infections (65-66). Therefore, conventional PCR and other molecular methods should be improved in order to cover these new mutant types or to revise the screening methods by implementing other recommended molecular techniques for HPV detection and typing (61,65,66). At present, valid genotyping procedures available commercially and in-house methods are approved in WHO proficiency testing, and could be considered as the best practical way for primary HPV testing in cervical cancer screening plans (10,66). The most important problems of HPV screening procedures, which need to be considered, are false positive and negative results, sampling preparation and shipment specimens. However, the most important objective is HPV diagnosis in early stages of cervical cancer and precancerous lesions and other related cancers and infections (67-71). Unfortunately, our search showed no special attention on the majority of home brew methods to optimize the test in applied conditions, regarding detection limit, frequency, and false negative and positive results, except a few studies (12). Another missed point in these papers was ignoring HPV multiple infections that were underlined in reports applying standard and approved methods (4,72-78). We did not expect a high rate of multiple infections in papers on normal populations attending gynaecology clinics, since it can be varied in each population. However, patients with cervical cancer or high-risk groups are at increased risk of infection with more than one type. The released data also showed that proper screening programs were almost missing to deprive females for early detection of cytological changes. The WHO reports on human papillomavirus and related cancers in Iran and its neighbouring countries showed that no comprehensive data are available from cancer registry on the incidence of cervical cancer, or age standardized incidence rate by histological types among females in this region (77).

Looking through these investigations and reported articles revealed some gaps, and lack of HPV strategy in policy makers of cervical cancer screening. Therefore, it seems that there is no HPV DNA primary testing plan for patients with cervical cancer in the countries of this region. This may be due to cost of the tests or lack of diagnostic surveillance system algorithm. Hence, the following suggestions can help the authorities and national policy-makers to overcome cancers related to HPV rate:

1. Commercializing the most popular molecular protocols in both screening and genotyping methods in order to reduce the sampling cost per test for health services and clinical laboratories.
2. Simultaneously performing co-testing of pap tests with molecular HPV testing for any screening of HPV infections.
3. Defining and designing an algorithm for care HPV, screening and prophylactic of cervical cancer programs. The designed algorithm can be different for any country, depending on the prevalence of most carcinogenic genotypes, environmental conditions and other parameters.
Conclusion
Further planning for regular HPV screening and genotyping by authoritative methods and development of favorable national vaccination program is necessary to prevent incidence of cervical carcinoma in the future.

Conflict of Interests
The authors declare that there was no conflict of interest.

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