# FOGSI GCPR on Screening and Treatment of Preinvasive Lesions of Cervix & HPV vaccination

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# Section 1: Screening and Treatment of Preinvasive Lesions of Cervix

## 1. Introduction
### 1.1 Background:

Cervical cancer is a major public health problem in India with an incidence of 1,22,844 cases and mortality of 67,477 cases every year[1,2,3]. Carcinoma cervix is the second most common gynecological malignancy amongst Indian women aged 25-44 years with an incidence of 3.5% after carcinoma breast (28.6%)[3,4]. The age standardized incidence rate for carcinoma cervix in Indian women is 22 per 100,000 women per year, which is the highest in South Asia compared to 19.2 in Bangladesh and 13 in Sri Lanka. Cervical cancer mortality is 12.43/100,000 per year[1-3]. Due to lack of an organized cervical screening program, the disease burden is high in India[5]. There is an intense need to standardize the diagnosis and management of pre invasive disease of cervix.

India is a vast country with diversity in social, cultural and religious practices, and this leads to differences in clinical practice and access to medical care with respect to screening and prevention of cervical carcinoma. Screening and treatment facilities are not available uniformly in the country and there are areas with shortage of gynecologists, pathologists, laboratories and colposcopists, thus limiting the establishment of an effective screening program. Therefore there is a need to develop good clinical practice recommendations (GCPR) that are evidence based and applicable to all possible clinical situations. Every screening program needs to incorporate adequate treatment and follow up of each screen positive case. The emphasis of these recommendations remains accurate detection followed by optimum management of pre invasive lesions of cervix.

These GCPR are unique as they have taken into consideration the available resources, clinical conditions, population preferences and research evidence in the Indian context while formulating the management algorithms. The FOGSI GCPR were developed by a panel of experts from all over the country. The experts reviewed the research evidence and met to formulate the recommendations. After brainstorming sessions, a

draft was prepared and once again critically reviewed by all contributors. After incorporating all the suggestion the final document was prepared.

## 1.2 Resource stratification:

FOGSI GCPR gives the option of using the modality that is available to the clinicians, in any part of the country and also provides the comprehensive overview of follow up and treatment if anything abnormal is detected. Various guidelines on screening and treatment of precursor lesions in all resource settings have been published by the World Health Organization (WHO, 2014)[6], Ministry of Health and Family Welfare (MOHFW, 2016)[7] and the American Society of Clinical Oncology (ASCO, 2016)[8]. FOGSI GCPR stratifies the healthcare systems into two major categories: settings with good resources and settings with limited resources. On the basis of resources available and individual preferences, one can choose the screening and treatment modality. In good resource settings, there is no lack of facilities including trained doctors, laboratories and equipment. Limited resource settings may lack adequate equipment, technology or staff to perform and interpret the results. The suggested modalities for screening and triage in each setting are shown in Table 1.

**Table 1: Resource stratification and suggested modalities for screening and management of precancerous lesions of cervix**

| Setting | Screening tools | Triage | Management options | Single visit approach Strategy |
|---|---|---|---|---|
| Good Resource Settings | Primary HPV DNA test or Co-testing (HPV test + Cytology) or Cytology or VIA | Cytology $\pm$ Newer modalities,* HPV test, HPV Genotyping for 16/18, Colposcopy, VIA | LEEP, Conization, Cryotherapy, Thermocoagulation | See and Treat approach |
| Limited resource Settings** | VIA | Colposcopy if available | Cryotherapy LEEP $\pm$ Conization, Thermocoagulation | Screen and treat Or Screen, See and Treat approach |

* Newer modalities (p16, ki 67 testing, mRNA testing, E6,E7 protein testing)

** Affordable HPV test (if available), including self-sampling, can be used.

Studies were reviewed and evaluated for quality according to the method outlined by the U.S. Preventive Services Task Force. The strength of recommendations was given to each good clinical practice recommendation as per the criteria endorsed by USPSTF in Table 2.

Table 2 : Strength of Recommendation[9]

| Quality of Evidence | Definition |
|---|---|
| I | Evidence obtained from at least one properly designed randomized controlled trial |
| II-1 | Evidence obtained from well-designed controlled trials without randomization |
| II-2 | Evidence obtained from well-designed cohort or case–control analytic studies, preferably from more than one center or research group |
| II-3 | Evidence obtained from multiple time series with or without the intervention. Dramatic results in uncontrolled experiments also could be regarded as this type of evidence |
| III | Opinions of respected authorities, based on clinical experience, descriptive studies, or reports of expert committees |
| Based on the highest level of evidence found in the data, recommendations are provided and graded according to the following categories: | |
| Level A | Recommendations are based on good and consistent scientific evidence |
| Level B | Recommendations are based on limited or inconsistent scientific evidence |
| Level C | Recommendations are based primarily on consensus and expert opinion |

**1.3 Single visit approach**

Successful implementation of cervical cancer prevention programs requires good linkages between screening and treatment. In the multiple visit approach there is a high incidence of loss to follow up and failure of compliance with recommended treatment. The single visit approach was initially tried in colposcopy clinics (See and Treat approach) but a similar strategy where visual inspection with acetic acid (VIA) and cryotherapy are performed in the same visit (Screen and Treat Approach) has been found to be effective, feasible and cost-effective for resource constraint settings. Single round of screening in women > 35 years has led to 25% reduction in life time risk of cervical cancer.[10,11] The suggested criteria for Single visit approach are shown in Table 3.

**Table 3 : Criteria for Single visit approach**

| See and Treat | Screen and Treat |
|---|---|
| In Colposcopy clinics | In Public Health Programs |
| Patient referred with abnormal cytology report | VIA detects abnormal lesion |
| Colposcopy scoring indicates a high grade lesion | Criteria for ablation fulfilled |
| Simultaneous treatment done – excision or ablation | Treat immediately, with or without biopsy |
| Low probability of over-treatment because high specificity of cytology | Lower probability of over-treatment in high prevalence areas |
| Post-hoc analysis of biopsy report/excision specimen | Post-hoc analysis is possible if biopsy was taken |

**See and Treat approach**

In a study done to evaluate the over-treatment rate when choosing a see and treat approach in patients with deviant cervical smear test results, 723 patients were analysed. High grade CIN was found in 70.3% of patients with LSIL showing that 29.7% of patients will be over treated with a See and Treat approach. For patients with HSIL, the rates of overtreatment were 6.7%[12]. Moreover See and Treat was more useful in women with ASC-H and HSIL cytology as immediate linkage to treatment led to better compliance and lesser chances of missing the lesions at biopsy[13].

**Screen, See and Treat approach**

This strategy was associated with 66% defaulter rates when patients were referred to other centers for colposcopy, but when colposcopy was done on the same site 97% women underwent the procedure and received optimum treatment. However a higher incidence of overtreatment was observed[14].

WHO has recommended evidence based guidelines for treatment of CIN 2,3 and screen and treat strategies to prevent cervical cancer. Depending on the availability of resources a strategy of screen with an HPV test and treat with cryotherapy or a strategy to screen with VIA and treat with cryotherapy or LEEP is recommended. The same expert panel has suggested that a strategy to screen with HPV and treat with cryotherapy or LEEP is preferred over a strategy of screen with cytology or HPV test followed by colposcopy and treat with cryotherapy (Screen, See and Treat)[15].

**1.4 How to choose an appropriate screening modality**

The three main screening modalities are human papillomavirus (HPV) testing, cytology (Pap smear) and VIA (Visual inspection by acetic acid). FOGSI recommends the use of HPV testing as the best method for cervical cancer screening, alone or in combination with cytology. However, centers with an established cytology program with good quality indicators may continue to do the same. VIA is a test with sensitivity comparable to cytology and suitable for use in low resource settings. Wherever possible, colposcopy should be used to obtain a guided biopsy. However, in its absence, the biopsy can be guided by VIA. In certain low resource settings, small low grade lesions may be considered for screen and treat approach without biopsy confirmation. Figure 1 depicts the algorithm to help clinician choose the most appropriate screening modality applicable to their health care setting. Table 4 summarizes FOGSI recommendations for screening in different resource situations. Each of these has been further discussed in detail below
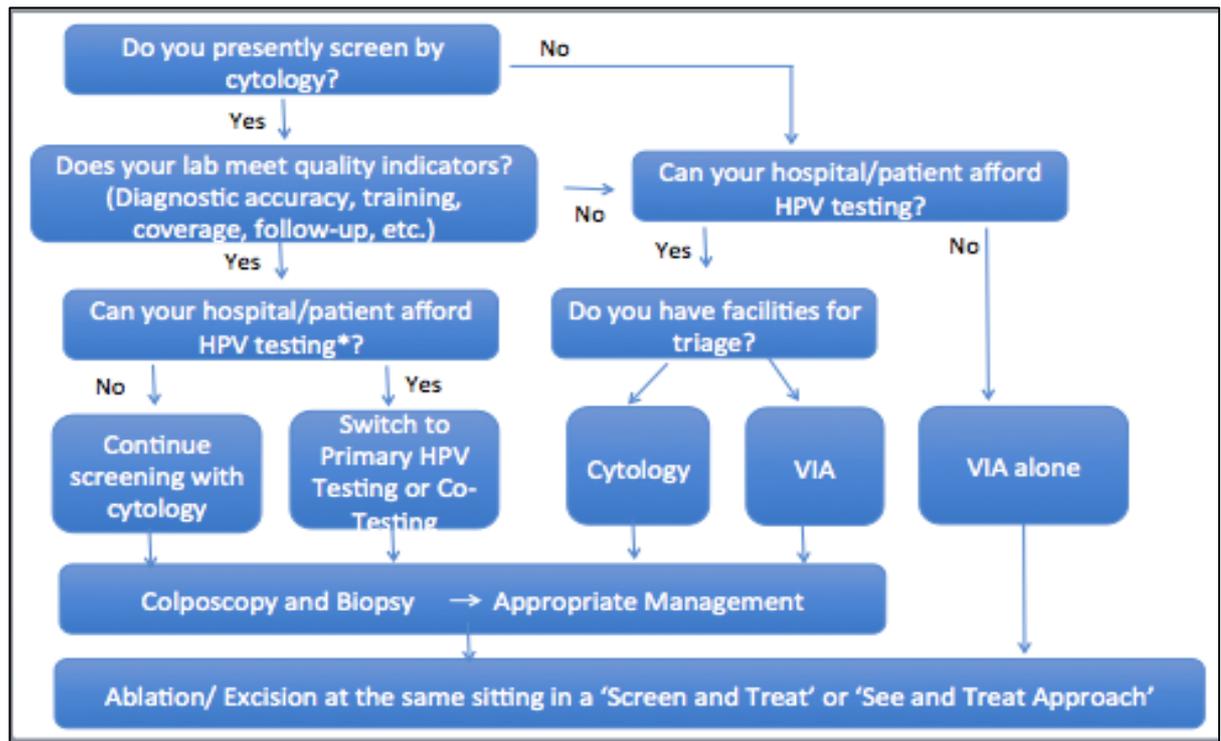
*Adapted from WHO[6]*

*Fig 1:* **Algorithm to help clinicians select the most appropriate screening modality based on resources**

**Table 4: Summary of FOGSI Resource-based Cervical Cancer Screening Recommendations**

|  | Good Resource Settings | Limited Resource Settings |
|---|---|---|
| **Modality** | **HPV testing**<br>• **Primary HPV testing**<br>• **Co-testing (HPV & Cytology)**<br>**Cytology**<br>**Colposcopy and biopsy**<br>**VIA** | **VIA**<br>**Colposcopy ± Biopsy** |
| **Target Age Group (years)** | **25-65** | **30 – 65**<br>**(N.B.: In postmenopausal women, screening with VIA may not be as effective)** |
| **Age to start (years)** | **Cytology / Primary HPV Testing at 25**<br>**Co-testing at 30** | **30** |

| Frequency | Primary HPV Testing or Co-testing - every 5 years Cytology – every 3 years | Every 5 years (at least 1-3 times in a lifetime) |
|---|---|---|
| Age to stop (years) | 65 with consistent negative results in last 15 years - women with no prior screening should undergo tests once at 65 years and, if negative, they should exit screening. | |
| Follow up after treatment (Method and Interval) | HPV testing (preferred) *or* Cytology 12 months | VIA 12 months |
| Screening following abnormal reports ≥ CIN 2+, irrespective of method of treatment | 20 years | |
| Screening in hysterectomized women | • Following hysterectomy in which cervix was removed for benign causes : no need for screening, unless there is history of previous cervical intraepithelial neoplasia <br> • Absence of cervix must be confirmed by clinical records or examination <br> • If indications for hysterectomy unclear, screening may be performed at clinician's discretion | |
| Follow up in women with CIN in hysterectomy HPE report | • Need to be screened with HPV at 6 months and 18 months | |

## 2.1 Age to start screening

In India, the burden of cervical cancer under the age of 30 years is very low and under 25 years is negligible. Women under the age of 25 should therefore not be screened regardless of the age of sexual initiation or other risk factors unless they present with symptoms[16]. Even in the West, the incidence of cervical cancer in women aged < 21 years has not changed with increased screening coverage over the last 4 decades[17]. Screening very young women is associated with relatively higher rates of un necessary evaluation and treatment of lesions that would otherwise regress[18].

**Recommendation:**

- In health care settings with good resources, screening should begin from age 25 years. (Level A)
- In health care settings with limited resources, screening can be delayed till age 30 years. (Level B)

**2.2 Screening periodicity:**

Women aged 30-65 years should preferably be screened with primary HPV testing or combined cytology and HPV testing ("co-testing") every 5 years. If the quality of HPV testing is of doubtful validity using non-standardized HPV testing methods, co-testing is to be preferred to primary HPV testing alone till enough data accrues. In case HPV testing is not available, cytology alone/VIA is an acceptable modality and should be repeated every 3 years[19]. Liquid based cytology does not improve the sensitivity over conventional cytology though it does decrease the rates of unsatisfactory smears and also allows the same sample to be used for other tests including HPV testing, gonorrhea, chlamydia, etc. Annual screening has no advantage , rather leads to increased number of un necessary colposcopies and is not currently recommended for any category[20,21,22].

In low resource situations, Ministry of Health and family welfare recommends screening 5-yearly with VIA till age 65 years. It is expected that this group of women will receive screening at least one to three times in their lives. Hence screening with cytology every 3 years or HPV test every 5 years is optimal to achieve maximum benefits[23].

**Recommendation:**

- In health care settings with good resources, women aged 25-65 years should undergo cervical cancer screening preferably with primary HPV test/co-testing every 5 years or with cytology every 3 years. (Level A)
- In health care settings with limited resources, women aged 30-65 years should undergo cervical cancer screening with VIA every 5 years, at least one-three times in their lives. (Level B)

**2.3 Age to stop:**

If screening is continued to age 90 years, the chances to prevent cervical cancer was only 1.6 cases and 0.5 deaths per 1000 women. Moreover it will lead to 58 extra false positives, 127 preventable colposcopies, 13 extra CIN cases undergoing treatment and

adding life expectancy by only one year per 1000 women.[24] Therefore women over 65 years of age with previous adequate negative screening and no history of CIN2+ within the last 20 years need not be screened[24]. Adequate negative prior screening means three consecutive negative cytology results or 2 consecutive negative co-tests within the 10 years before ceasing screening, with the last test within the last 5 years. In women who have undergone hysterectomy with a report of CIN2+ lesions, screening should be continued for 20 years from the age of surgery.

**Recommendations:**

- Regardless of resource situation, women should continue to be screened for cervical cancer till age 65 years. Thereafter screening can be discontinued in women with consistent negative results in last 15 years. (Level A)

- In women who have undergone hysterectomy for benign reasons screening is not advised, unless there is a history of previous cervical intraepithelial neoplasia. (Level A) Absence of a cervix must be confirmed by clinical records or examination, however if the indication for hysterectomy is not clear, screening may be performed at clinician's discretion.

- In women who have undergone any modality of treatment for CIN 2+, screening for 20 years following the time of treatment recommended. (Level A)

**3. Screening using available modalities**

**3.1 HPV test as primary screening modality**

**Primary HPV testing**

HPV is the main causal factor for the development of cervical cancer. Eighteen HPV subtypes including HPV16 and HPV18 are known to be associated with invasive cancer[25]. HPV testing alone for primary screening is recommended in women aged 30 years and older, as a negative HPV test provides greater reassurance against CIN3+ in the subsequent 5 to 7 years than cytology alone and is nearly as reassuring as a negative co-test.

The cumulative incidence rates of CIN3 and cancer in HPV negative women aged > 25 years was reported as 0.34% (95% CI, 0.10–0.65) and in cytology negative women was 0.78% (95% CI, 0.53–1.09)[26]. Three year CIR of CIn3+ in women who were HPV negative and cytology negative was 0.30% (95% CI, 0.05–0.62). Although the

primary hrHPV screening detected approximately 50% more CIN3+ compared to cytology, it also resulted in higher number of colposcopies.

Ronco et al.[27] analyzed follow-up data from 4 published RCTs of hrHPV based screening including the NTCC (Italy), ARTISTIC (United Kingdom), Swede screen (Sweden), and POBASCAM (Netherlands) trials. There was no difference in cancer detection in the first 2.5 years, but after extended follow-up, the incidence of invasive cervical cancer was significantly lower in women initially screened with hrHPV based testing compared to cytology alone test. (RR, 0.45; 95% CI, 0.25– 0.81).

The benefit of using HPV testing as primary screening method is its high sensitivity and negative predictive value, and this allows safe prolongation of the screening intervals[28]. HPV testing is the recommended primary screening method in women aged $\geq$ 30 years. Recently, the Cobas test (Roche) which detects hrHPV, and genotypes HPV 16/18, has been approved for primary HPV testing in women aged >25 years[9]. HPV testing is a preferred method of co-testing in women >30years or as primary screening at age >25 years depending on the test used[29]. It is useful in triaging women with ASCUS cytology[30] and following treatment for CIN. It is more sensitive but less specific than cytology in post treatment follow-up and this leads to earlier diagnosis of persistent or recurrent disease[31].

It is hoped that in time the development of affordable, point of care HPV tests will permit the use of HPV testing in limited resource settings also.
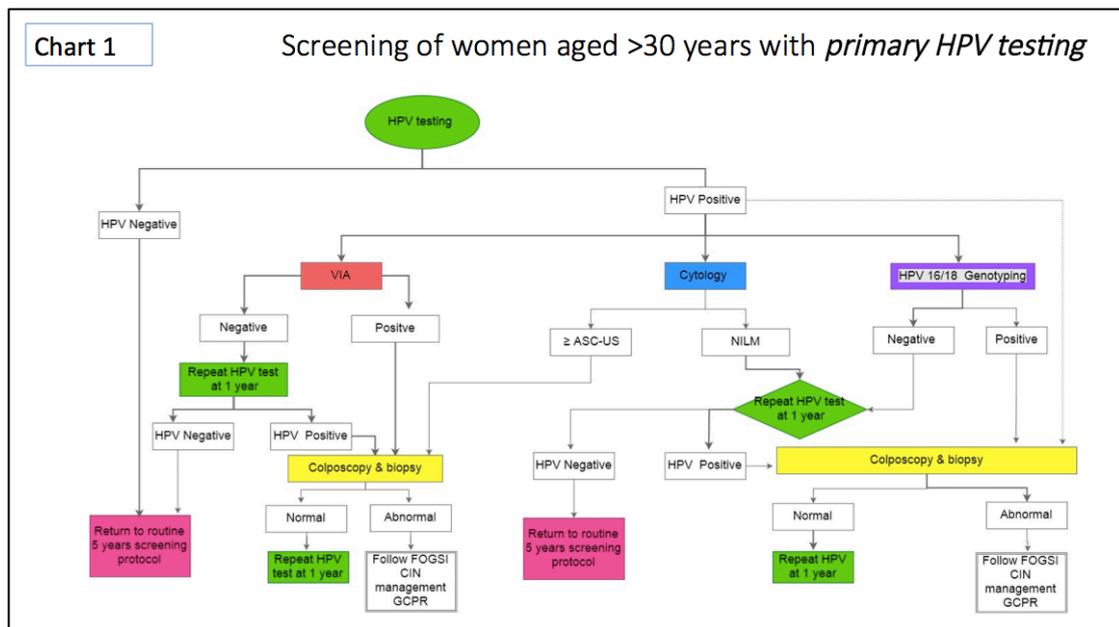


**Chart 1: Screening of women aged > 30 years with primary HPV testing**

**Recommendations**

**Presently, HPV testing is available only in good resource settings.**

1. For women aged 30-64 years, primary HPV testing is acceptable screening method. (Level A)

2. Women with negative HPV test, should return to next screening after 5 years. (Level A)

3. Women with positive HPV test should be triaged either with cytology, HPV genotyping or VIA depending on availability. (Level B)

> a. When HPV positive women are triaged with cytology, a normal cytology report should be followed by repeat HPV test at 1 year. If cytology report is ≥ASCUS colposcopy and directed biopsy should be advised. (Level A)

> b. When HPV positive women are triaged with HPV genotyping 16, 18, a positive result should be managed with colposcopy and directed biopsy and further follow up should be done as per CIN management charts. If HPV genotyping is negative in an otherwise HPV positive woman then one can offer cytology (Level B)

> c. When HPV positive women are triaged with VIA, a negative VIA should be followed by repeat HPV test at 1 year. On the other hand if VIA is positive, colposcopy and directed biopsy should be advised. (Level B) Further follow up and management should be done as per FOGSI CIN management chart.

**Choice of HPV test:**

HPV tests detect high-risk HPV types, either by amplification of a viral DNA fragment (with or without genotyping), or through mRNA identification.

HPV DNA tests identify the DNA of one or more oncogenic HPV types with direct genomic detection or by amplification of a viral DNA fragment using polymerase chain reaction (PCR) to obtain copies, both conventionally and in real-time. HPV genotyping identifies specific viral types (usually HPV 16 and 18). The mRNA tests detect expression of HPV E6 and E7 onco proteins.

**Table 6 : HPV tests used for cervical cancer screening**

| Test | Technique | Name |
|------|-----------|------|
| DNA | Direct Genome detection | Hybrid Capture 2 |
| | | *care*HPV test |
| | Amplification | GP5+/GP6+ bio PCR-EIA |

| | | Cervista HPV HR |
|---|---|---|
| | Amplification and genotyping of HPV-16 and HPV-18 | Cervista HPV 16/18 |
| | | Cobas HPV test |
| | | Xpert HPV |
| | | Abbott Real time high risk HPV assay |
| | | Papillo check |
| RNA | Amplification of E6/E7 proteins | Aptima HPV assay |
| | | PreTect HPV-Proofer HPV |
| | Monoclonal antibodody | AVantage HPV E6 Test |

The U.S. Food and Drug Administration (FDA) in 1997 approved Digene Hybrid capture (HC) HPV DNA for triaging women with equivocal cytology report. In 2003 HC2 came as an adjunctive screen with cervical cytology for women 30 years and older. HC2 has been the gold standard for cervical cancer screening until 2011-2012 when FDA approved Aptima, Cervista and Cobas HPV assay.

HPV testing can not differentiate between persistent and transient infection, therefore it has 3%–4% lower specificity than cytology (at cutoff ASC-US+).[32] hrHPV detection methods comprise of HPV DNA assays and E6/E7 mRNA assays. Hybrid capture II and G5+/6+are clinically validated prototype assay as they have shown better performance to lower the incidence of CIN3+.

**3.2 Co testing as primary screening modality**

For women aged 30–65 years, co-testing with combined cytology and HPV testing every 5 years is the preferred method of screening presently. Although there has been widespread endorsement of primary HPV testing.[9] FOGSI perceives the need for more Indian data on the validity of primary HPV testing before this is endorsed as the preferred method in India. The negative predictive value is high when both test results negative and there is very high level of reassurance that they will not be at risk for cervical cancer for a long time. Thus incorporation of HPV testing with cytology has the potential to reduce the incidence and mortality of cervical cancer in women aged 30 years and older. [33,34,35]

Co-testing leads to earlier diagnosis of CIN 3+ and Cancer.[4] Incorporating HPV with cytology helps in finding more AIS than cytology alone and negative co-test allows spacing screening in every five years[27,37]. Co-testing has been successfully implemented in developed countries like United States and recent data show a continued decline in cervical cancer rates.

Studies have also shown that incorporating HPV testing detects more AIS than cytology alone. The meta analysis of four large RCTs [38,39] namely NTCC phase1, POBASCAM, Swede screen and ARTISTIC trial, compared co testing with cytology screening alone in European women aged 30 to 64 years. The cumulative CIN3+ detection after two rounds of screening was similar between co-testing and cytology group. [38,39,40]

The patients with negative co testing have lower incidence of cervical cancer.[41] The 5-year cumulative incidence of CIN 3 or cancer was less than half the risk associated with negative cytology alone.[41] This is the basis of extending the screening interval from every 3 years to every 5 years with the use of co testing.[37]

While HPV testing is more sensitive than cervical cytology testing, it is somewhat less specific. Co-testing in women younger than 30 years would lead to an increase in unnecessary workup procedures without a corresponding decrease in cervical carcinoma incidence because of increased probability of clearance of infection by immune system.[42]

Dillner and colleagues[34] using pooled data from European studies showed a greater reduction in CIN3+ disease in co-testing cohort than Primary HPV screening cohort. Co testing was more specific than primary HPV at 6 years follow up as CIN3+ disease was found in 24% fewer women in co-test negative cohort than negative primary HPV cohort. Similarly other studies have also confirmed that co-testing with Pap+ HPV identified more cervical pre cancers and invasive cancers than the HPV or Pap test alone.[43,44] Moreover co- testing using highly specific HPV assay was clinically better and more   cost effective as compared to Primary HPV testing.[45]

The older version of United States Preventive Services Task Force (USPSTF)[36] organizations endorsed concurrent Pap and HPV testing ("co-testing") for women age 30 and older in the cervical cancer screening guidelines[28] but the latest guidelines recommend primary HPV screening as the preferred modality over co testing.  The women who test HPV-negative/Pap-negative have enough safety against cervical cancer that they can return for routine screening in 5 years. However, in India there is

limited availability of HPV testing and often the tests used are not standardized. Hence it is preferable at the present time to recommend co-testing until sufficient Indian data accrues.
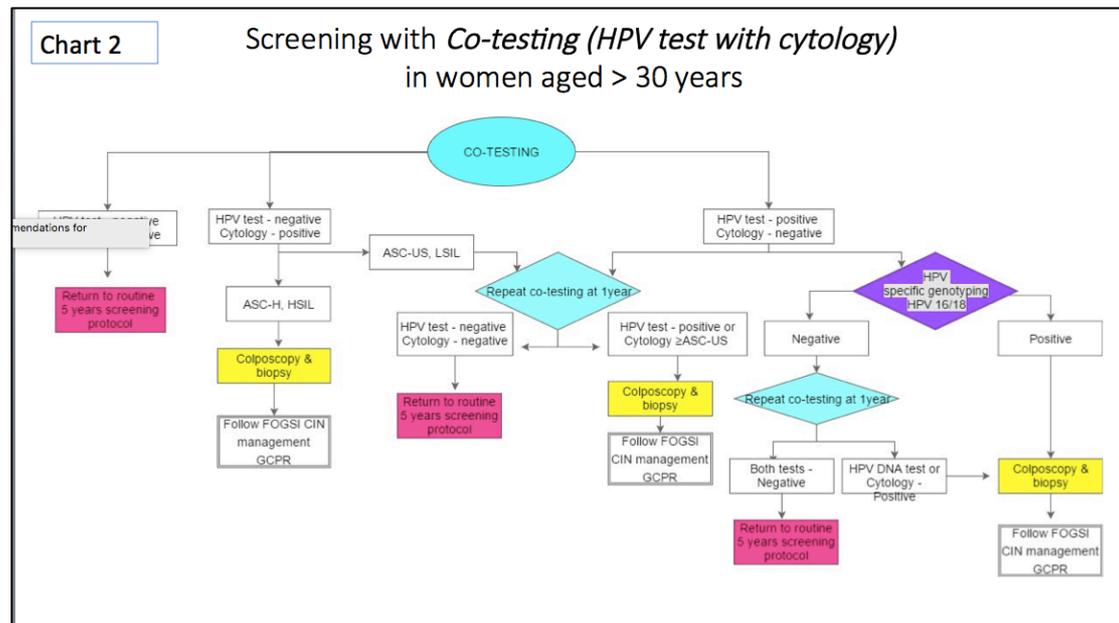


**Fig 2 : Screening with *Co-testing (HPV test with cytology)* in women aged > 30 years**

 **Recommendations** (Chart 2)

1. For women aged 30-64 years, co-testing is the preferred screening method if resources are available (Level A)

2. The women with negative cytology and HPV test, should return to routine screening with co-testing at 5 years. (Level A)

3. The women with positive cytology and negative HPV test, the follow up depends on cytology test.

    a. If cytology is ASCUS or LSIL, repeat co-testing is advised at 1 year. (Level B)

    b. Women with cytology ≥ ASCUS or HPV test positive on repeat co-testing, should be managed by colposcopy and directed biopsy. (Level A)

    c. Women with ASC-H or HSIL on cytology and HPV test negative, should be managed by colposcopy and directed biopsy. (Level A)

4. The women with HPV test positive and cytology negative, should either go for repeat co-testing at 1 year or should go for HPV specific genotyping. (Level C)

    a. Women who go for HPV genotyping, if result is negative should be followed by repeat co-testing at 1 year and any abnormal test results should be managed as per FOGSI GCPR recommendation. (Level B)

    b. If the HPV genotype result is positive, should be managed by colposcopy and directed biopsy. (Level A).

## 3.3 Cytology as the primary screening modality

Cervical cytology is the most commonly used screening test for detecting cervical preinvasive and invasive lesions of cervix. Terminology for reporting cervical cytology was standardized by the Bethesda System in 1988 and was revised last in 2014[46].

It is important to highlight that the results of cervical cytology cannot be used to make a definitive diagnosis or start treatment except when it is HSIL. Rather the results are used to guide further evaluation with colposcopy and or biopsy, and treatment decisions are based upon diagnostic results from such examination.

### 3.3.1 Terminology for squamous cell abnormalities [46]

**Interpretation of cervical cytology reports**

A. Squamous cell abnormalities in the cytology report can be interpreted as CSIL ( Cervical squamous intra epithelial lesions ) which are further stratified into

- **Low grade squamous intraepithelial lesion (LSIL)**: LSIL in young women is a transient HPV infection and has relatively lesser risk of cancer progression. This is associated with probable histologic diagnosis of CIN 1.
- **High grade squamous intraepithelial lesion (HSIL)**: This is associated with probable histologic diagnosis of CIN 2/3.
- **Atypical squamous cells**
  - ASCUS
  - ASC-H : LSIL cervical cytologic specimens that contain a few cells that are suspicious for but not diagnostic of HSIL are reported as atypical squamous cells, cannot exclude a high

grade squamous intraepithelial lesion (ASC-H). Approximately 5 to 10 percent of ASC results are designated ASC-H.

B. **Glandular cell abnormalities**:

Benign appearing endometrial cells are reported only in women ≥45 years.

- **Atypical glandular cells (AGC):** Endocervical, endometrial, or not otherwise specified is noted. Upon further evaluation, either high grade squamous or glandular abnormalities are found in 10 to 39% of women with a finding of AGC on cytology.
- **Atypical glandular cells, favor neoplasia (AGC-FN)**: Endocervical or not otherwise specified is noted. This designation is for specimens that show features suggestive of but not sufficient for an interpretation of adenocarcinoma.
- **Endocervical adenocarcinoma in situ (AIS)**
- **Adenocarcinoma**

**3.3.2 Summary of rationale for management protocols**

**Risk of malignancy in age group 30-64 years**

The risk of progression of abnormality varies with each cytological abnormalities. (Table 7) Depending on the 5-year risk of progression to CIN lesion grade 3 or more follow up protocols are decided. (Table 8 )[47,48,49]

**Table 7: Risk of progression with cytological abnormalities**

| Cervical cytology | Incidence | | |
|---|---|---|---|
| | CIN 2+ | CIN3+ | Invasive |
| **NILM** | 0.68% | 0.26% | 0.025% |
| **LSIL** | | | |
| Only cytology | 16% | 5.2% | 6.1% |
| LSIL, HPV + | 19% | 6.1% | |
| LSIL HPV - | 5.1% | 2.0% | |
| **ASCUS** | | | |

| Only Cytology | 6.9% | 2.6% | 0.18% |
|---|---|---|---|
| HPV + | 18% | 6.8% | 0.41% |
| HPV - | 1.1% | 0.43% | |
| **ASC-H** | | | |
| Cytology alone | 35% | 18% | 2.6% |
| **HSIL** | 69% | 47% | 7.3% |
| **Atypical glandular cells** | 13% | 8.5% | 2.7% |
| **Squamous cell cancer** | 84% | 84% | 68% |

**Follow up protocols[50]** based on the 5 year risk of CIN lesion grade 3 or more severe the recommended protocol is given in Table 8

**Table 8 : Follow up protocols based on risk of progression**

| Risk of progression | Recommended modality |
|---|---|
| >5 % | Colposcopy |
| 2 to 5 % | Repeat testing in 6 to 12 month |
| 0.1 to 2 % | Repeat testing in three years |
| <0.1% | Repeat testing in five years (the same as routine screening) |

**Management of ASCUS in women aged 30-64 years**

The relative incidence of finding ASCUS in cervical cytology in this age group is approximately 2.8%[48]. Risk of having higher grade abnormalities in this age group varies depending upon HPV positivity as depicted in Table 7. The prevalence of HPV positivity with ASCUS cytology is 23-74%[51]. An ASCUS report on cytology can be triaged either with reflex HPV or repeat cytology or immediate colposcopy.

ASCUS/LSIL Triage Study (ALTS)[52] was a landmark randomized controlled trial that evaluated the strategies to manage ASCUS report. In this trial, 3488 women with ASCUS report were recruited to either immediate colposcopy, HPV triage, or repeat

cytology at six months interval. In the repeat cytology arm cytology was done every 6 months for two years and colposcopy was reserved for women with report of HSIL. The HPV triage group was found to have the highest sensitivity for detection of cumulative cases of CIN 3(72.3%). Conversely the immediate colposcopy and conservative management groups had similar sensitivity (54.6% and 55%). The rate of colposcopy referrals were lowest in the conservative management group (12%), followed by HPV triage (56%), and by definition, immediate colposcopy (100%). A single enrolment HPV test could detect 92.4% of the CIN grade 3 cases. Repeated cytology, would have required two visits to achieve similar performance (sensitivity (95.4%) and would have referred 67.1% to colposcopy even at an ASCUS threshold. Therefore, in women with ASCUS cytology, for detecting CIN 3, HPV triage is as sensitive as immediate colposcopy, and at the same time also reduces colposcopy referrals to half. On the other hand 6 monthly repeat cytology strategy, requires at least two follow up visits and more colposcopy examinations than HPV triage. However, the main limitation of this trial is that it was conducted in 2001 and at that time ASCUS category also included ASC-H. Thus the background rate of HPV infection and CIN 3 was relatively higher.

Cost effectiveness of triage of ASCUS with reflex HPV test has been found as the most cost effective strategy as compared to immediate colposcopy, repeat cytology, and two visit HPV testing[54,55]. Furthermore HPV genotyping would not result in a change in management from a positive HPV test, but would add cost. Conversely there is a 15-33% risk of missing a significant abnormality if one relies on a single repeat smear, and also there is a need for multiple follow up visits, delay in histological diagnosis and also lack of protocols about optimum frequency of testing[56,57]. In women with persistent HPV infection but normal colposcopy, the risk of developing invasive disease is higher than those with transient infection.

The five-year risk of CIN3+ in women with single positive test and negative cytology was 4.5%, but it increased to7.4% in those with persistent HPV positive cases. Similarly, another study observed the five-year risk of CIN 3+ with a single HPV positive test and negative cytology as 2% and it increased to 8% in women with two positive results.[59] This explains the rationale behind doing co testing in such situations

at 1 year. Conversely the 5 year risk of CIN3+ was similar in both HPV negative ASCUS (0.43%) and negative cytology regardless of HPV status(0.26%), however the risk in those with negative cytology and negative HPV testing was minimal (0.08 percent)[48,60].
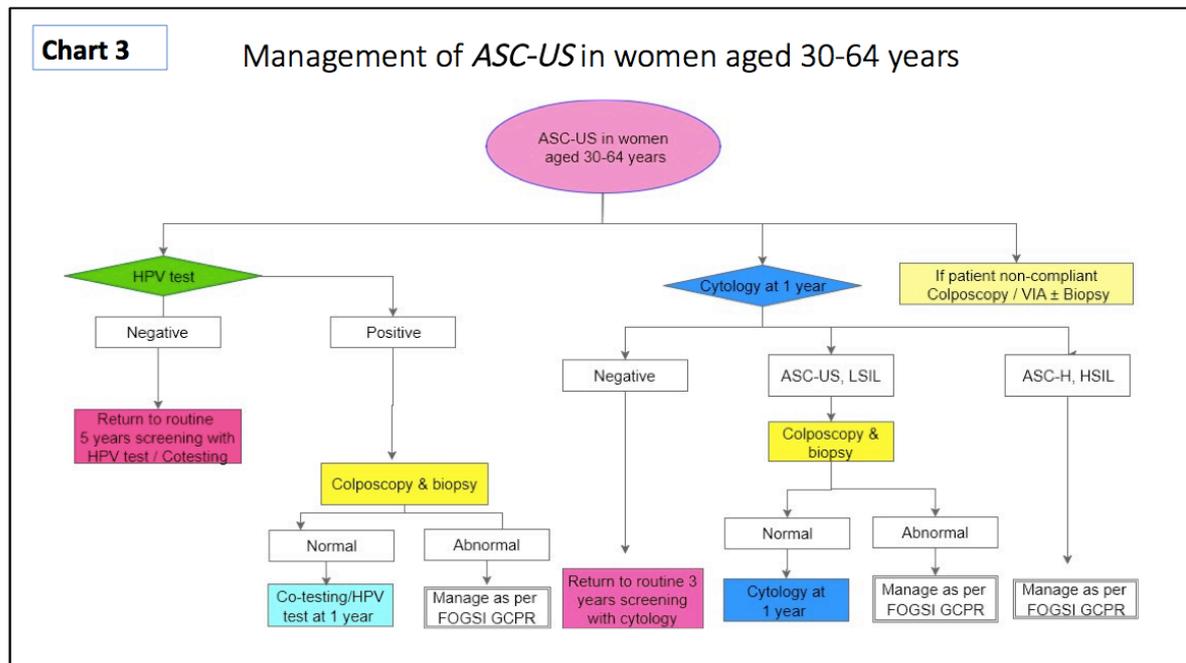


**Fig 4 : Management of *ASC-US* in women aged 30-64 years**

**Recommendation (Chart 3)**

1. Women aged 30-64 years with ASCUS cytology should be triaged preferably with HPV. (Level A)

2.The women with ASCUS cytology and positive HPV test should be managed by colposcopy and directed biopsy (Level A)

3. The women with ASCUS cytology and negative HPV test should be followed with co testing at 5 years. ( Level B)

4. In women with HPV positive ASCUS, normal colposcopy result should be followed by repeat co testing at 1 year and any abnormal test results should be managed as per FOGSI GCPR recommendation. (Level B)

5.Women aged 30-64 years with ASCUS cytology can also be triaged with repeat cytology at 1 year if facilities for HPV not available. In situations where patient is noncompliant then depending on the availability one can either do Colposcopy or VIA followed by biopsy if suspicious areas identified. (Level C)

4. In women with ASCUS cytology report, triaged with repeat cytology at 1 year, If repeat cytology is negative then patient should return to routine screening with cytology at 3 years. (Level B)

5. In women with ASCUS cytology report, triaged with repeat cytology at 1 year, If repeat cytology is ASCUS or LSIL, colposcopy and directed biopsy should be done and if that is normal, a cytology should be repeated after 1 year. The abnormal colposcopy results should be managed as per FOGSI GCPR chart (Level B)

6. The cytology report showing ASC-H or HSIL should be managed as per FOGSI GCPR.

**Management of ASCUS or LSIL cytology in women aged < 30 years**

In this age group the incidence of cervical cancer is 1.4/100,000 women but the incidence of HPV infection in women <40 years with LSIL or ASCUS cytology is as high as 40.4 percent[50]. Therefore, HPV testing is not of much relevance in this age group. Moreover 70% of CIN 2 lesions regressed in younger women (mean age 20.4 years) and progression for CIN 3 was also as low as 0.5%. Apart from this the colposcopy and treatment may do more harms than benefits in younger women with respect to their obstetric performance. Therefore, colposcopy should be done in this age group only if cytology results are persistent or severe.
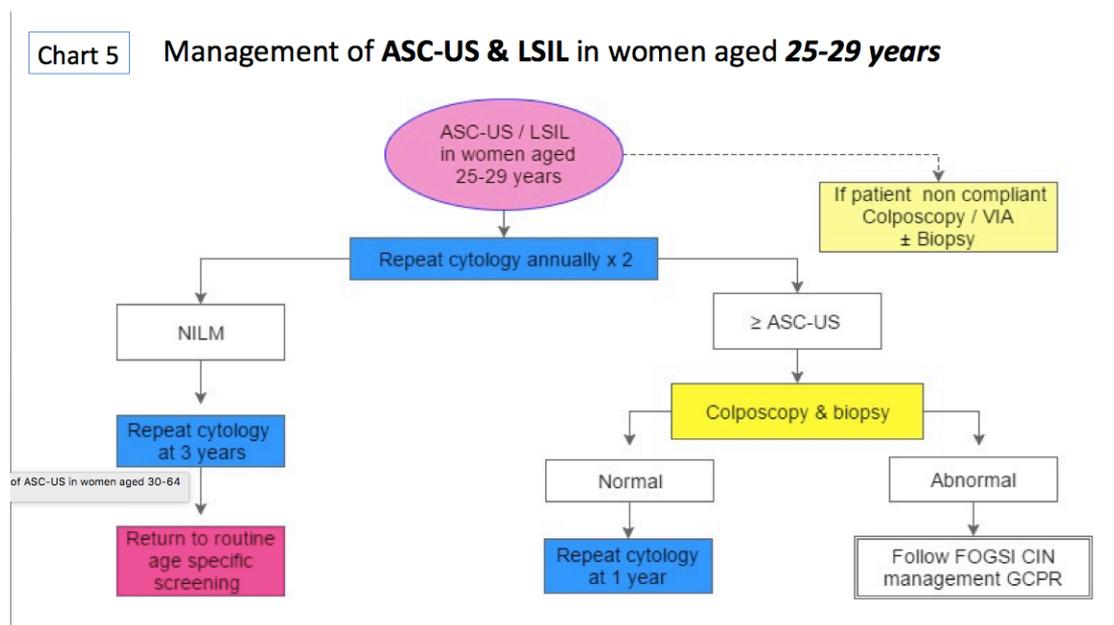


**Fig 5: Management of ASC-US & LSIL in women aged *25-29 years***

**Recommendations: (Chart 5)**

1. Cytology result showing ASCUS or LSIL in women aged 25-29 years HPV testing should not be done. (Level A)

2. These patients should be followed preferably with repeat cytology at 1 year twice, or if patient is non-compliant and facilities are not available then colposcopy or VIA is an acceptable modality. (Level C)

3. If repeat cytology is Negative for intraepithelial lesions or malignancy can return to routine age specific screening. (Level B)

4. If repeat cytology is ASCUS or higher colposcopy and directed biopsy should be done and if normal cytology should be repeated at 1 year. (Level B) If colposcopy is abnormal the patient should be managed as per FOGSI GCPR recommendations.

**Management of LSIL in women aged 30-64 years**

Women aged 30-64 years when screened with cytology, 0.97% will have report as low grade squamous intra epithelial lesions[61]. LSIL is associated with higher rates of HPV positivity. The risk of CIN 2+ and invasive disease with LSIL is shown in Table 7. Women in this age group with LSIL cytology report observed HPV positivity rates of 80% and the rate of decline of positivity was also lesser than younger women.[52] Therefore, majority of guidelines do not recommend the use of HPV triage with LSIL cytology, contrary to ASCUS cytology. The immediate colposcopy strategy for LSIL cytology had a sensitivity of 55.9% for detecting CIN 3 diagnosed over 2 years follow up. On the other hand, a conservative approach of doing repeat cytology and referring patients if reported as HSIL could detect 48.4% of CIN3 with an overall referral rate of 18.8%. Choosing lower grade cytology for referrals would increase the sensitivity but at the cost of higher referral rates. Therefore, it was concluded that LSIL cytology is best managed by colposcopy initially, because there was no useful triage strategy identified[52].

The strategy of immediate colposcopy in LSIL cohort was seen to associated with higher number of colposcopies and at least double rates of CIN 1detection than a strategy of repeating cytology (21 vs. 8%, RR: 2.58, 1.69–3.94) and this suggested that doing immediate colposcopy is likely to detect a higher number of insignificant lesions. Conversely the detection rate of CIN2 was higher in the immediate

colposcopy group at 18 months (6.5 vs. 3.2%, RR: 2.04, 1.52–2.73), but the differences were insignificant at 24 months of follow up (7.6 vs. 5.4%, RR: 1.45, 0.87–2.40). Thus it was concluded that incidence CIN2, CIN2+, CIN3+ was higher for immediate colposcopy than repeat cytology group, however the difference was not statistically significant may be due to smaller samples size. (CIN2: 8.4 vs. 11.6%, RR: 1.72, 0.66–4.48; CIN2+: 23.4 vs. 27.5%, RR: 1.43, 0.51–4.01, CIN3+: 15 vs. 15.9%, RR: 1.24, 0.39–3.94). Immediate colposcopy group had significantly higher detection rates of CIN3+ as compared to repeat cytology approach (30.9 vs. 17%, RR: 1.80, (1.11–2.92)[61].

**LSIL in women aged 25-29 years**

These women have a higher prevalence of HPV but at the same time there is increased chances of spontaneous regression with low risk of progressing into invasive disease. Thus the management in this age group is based solely on the cytology results[62,63].

**LSIL in postmenopausal age group**

Postmenopausal women with LSIL cytology should be triaged either with reflex HPV or repeat cytology at 6 and 12 months or colposcopy.

If HPV is positive, colposcopy is done but if HPV is negative then a repeat cytology should be done at 12 months. If two consecutive test reports are negative, then patient may resume routine screening. Similarly, if triaged with repeat cytology at 6 and 12 months if repeat test shows a result of ASCUS or higher colposcopy should be performed[64,65,66].

**Fig 6: Management of *LSIL* in women aged 30-64 years**

**Recommendation (chart 4,5)**

1. Women aged 30-64 years with cytology report LSIL should preferably undergo colposcopy and biopsy (Level A)

2. HPV triaging is acceptable in women aged 30-64 years with cytology report of LSIL (Level B)

**Management of women with ASC-H cytology (Atypical squamous cells cannot exclude high grade squamous intra epithelial lesion on cervical cytology)**

The probability of finding ASC –H in a cytology report is only 0.17%. The risk of invasive disease with this cytology is depicted in Table 7[48]. The rate of HPV positivity in women with ASC-H cytology is as high as 67%. Therefore, HPV testing is not required because it does not affect management of the patient. These women should be evaluated by colposcopy with special attention to adequate visualization of transformation zone and endo cervical curettage. However, if above conditions are not fulfilled a diagnostic excisional procedure should be done. This protocol is applicable in younger women with ASC-H also[50,67].

**Recommendations (chart 6)**

All women with ASC-H cytology should be evaluated by colposcopy. (Level A)

**Evaluation of HSIL**

The relative incidence of finding a cytology report as high grade squamous intra epithelial lesion is approximately 0.21%[48]. These women are at substantial risk of developing high grade CIN and invasive disease, therefore needs further evaluation. There is no role of HPV triage in this group because of high incidence of HPV positivity (89-97%).[68] The strategy for managing these women is to do either colposcopy or immediate LEEP [30,50]. While doing colposcopy, endocervical sampling is mandatory and if the colposcopy is unsatisfactory, except during pregnancy, a diagnostic excisional procedure should be the next step.   A negative colposcopy implies that the colposcopic examination is adequate and the endocervical curettage is negative.   Immediate cervical ablation is discouraged because ablative procedures do not provide a specimen for diagnostic evaluation.
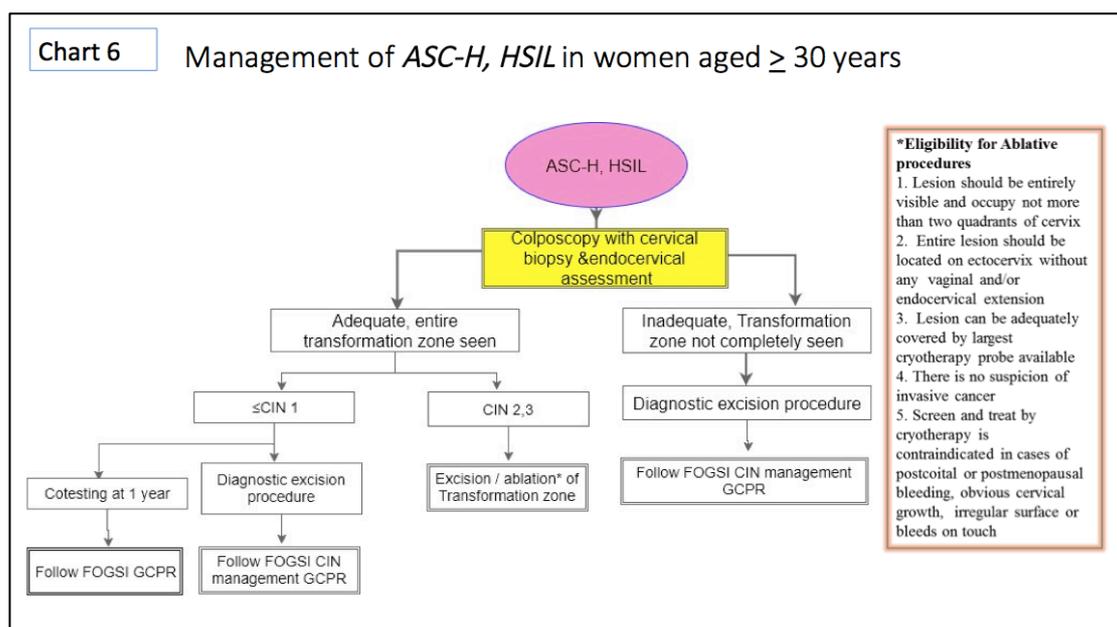


**Fig 7 : Management of *ASC-H, HSIL* in women aged ≥ 30 years**
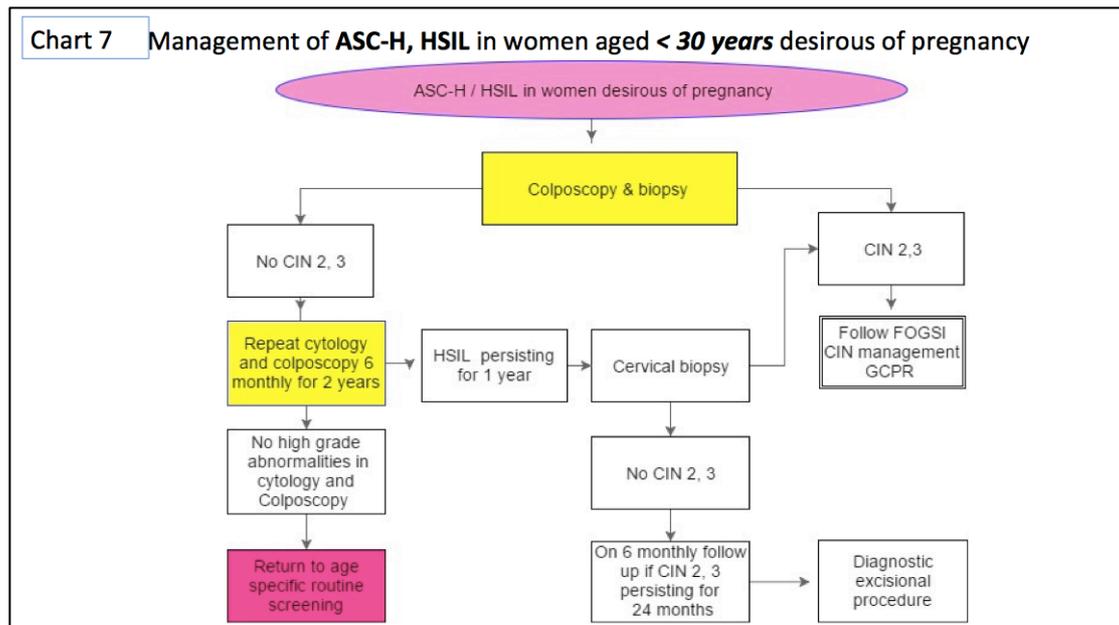
**Fig 8: Management of ASC-H, HSIL in women aged < *30 years* desirous of pregnancy**

**Recommendations (Chart 6,7)**

1. Women with cervical cytology report of HSIL should be advised to undergo colposcopy and directed biopsy. (Level A)

**Evaluation of atypical and glandular cells in cytology report**

Terminology [69,70]

**Atypical glandular cells:** The subcategories include

- o Endo cervical, endometrial or not otherwise specified (NOS).
- o (The term atypical glandular cells of un determined significance is no more existing now)
- o Atypical glandular cells favor neoplasia is the term used for specimens showing features suggestive of, but not sufficient for, interpretation of adenocarcinoma.
- o Endo cervical adenocarcinoma in situ (AIS)
- o Adenocarcinoma

When glandular cell abnormalities are present, it should be noted whether there are changes favoring neoplasia. The relative incidence of typical glandular cells on cervical cytology is approximately 0.1-2.1%.[71] These cells are associated with premalignant or malignant disease in 30% instances in women older than 40 years[72]. Even with atypical glandular cells majority of lesions are squamous and not glandular. The presence of atypical glandular cells on cytology is a significant marker for the presence of pre malignant disease and risk is known to increase with age[73]. Incidence of endometrial carcinoma was 12.7%, ovarian carcinoma 1.4%, and cervical adenocarcinoma was 0.9% in women more than 50 years with atypical glandular cells in cytology report. On the other hand, women 40-49 years with atypical glandular cells showed malignancy in only 2.8% cases and only 2% women <40 years showed malignant changes.[72] Thus it is mandatory to assess these women for both cervical as well as endometrial malignancy.

All women with any of the above mentioned AGC category except AGC-endometrial should be evaluated with colposcopy and directed cervical biopsy along with endo cervical sampling. Endometrial sampling should be done for women > 35 years' age and also younger women if high risk of developing endometrial malignancy. HPV testing is not required. Despite comprehensive evaluation if woman has negative findings then, possibility of other primary including ovarian, fallopian tube should be considered. A systematic review with 7000 women having a cytology report of atypical glandular cells, found that 6.4% had ovarian or fallopian tube carcinoma and 6.9% had cancers of sites other than the cervix, endometrium, ovaries, or fallopian tube[74]. These women should then be followed by a transvaginal ultrasound. Women with an adnexal mass are further evaluated for ovarian or tubal cancer.
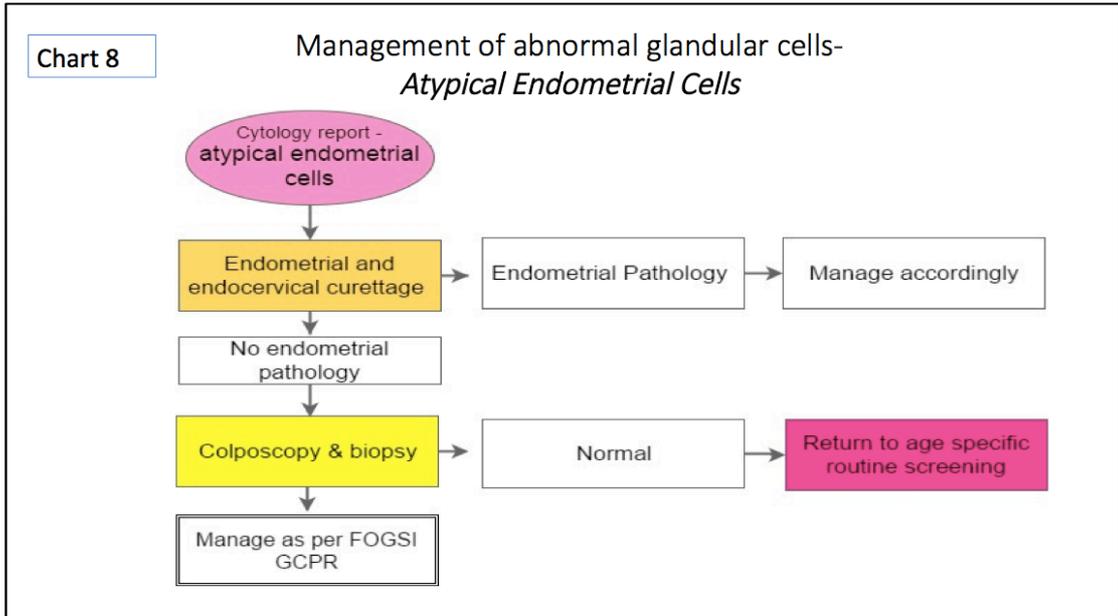
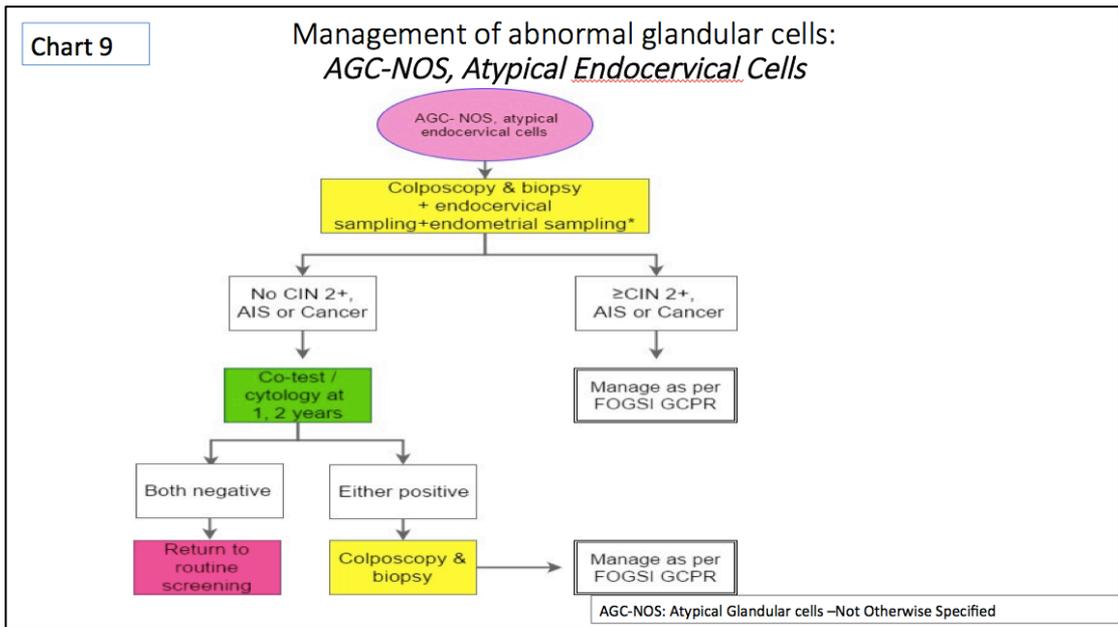**Fig 9: Management of abnormal glandular cells:***Atypical Endometrial Cells*



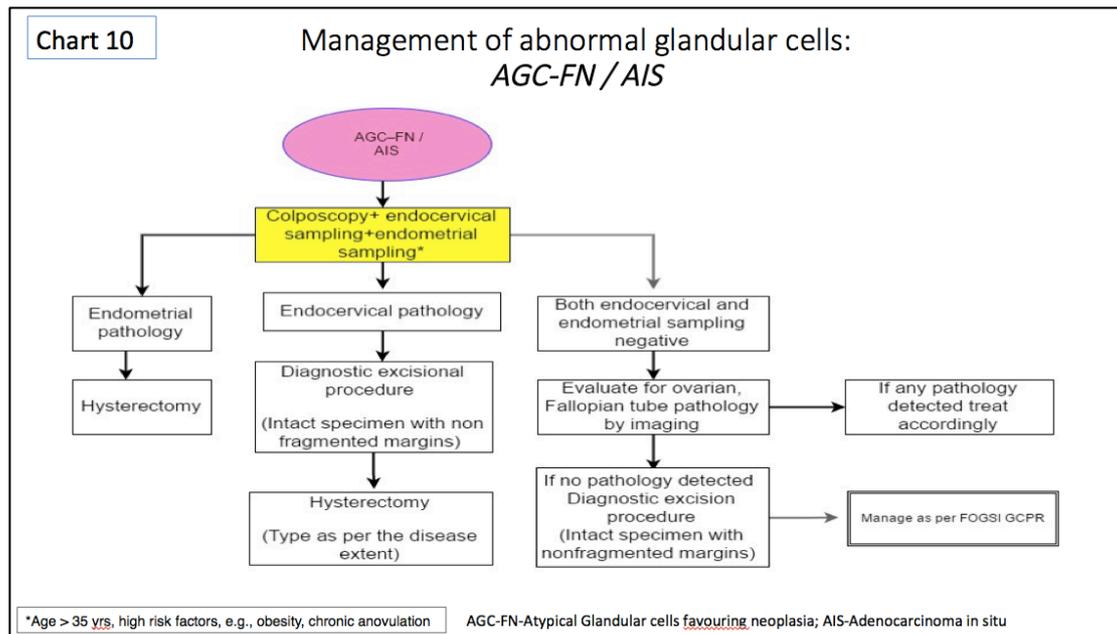**Fig10: Management of abnormal glandular cells:** *AGC-NOS, Atypical Endo cervical Cells*

**Fig 11:Management of abnormal glandular cells: AGC-FN / AIS**

**Recommendations (Chart 8,9) :**

1. Women with cytology report of atypical glandular cells should be evaluated with colposcopy and directed biopsy along with endo cervical and endometrial sampling. (Level A)

**3.4 VIA as primary screening modality**

Visual inspection of the cervix with acetic acid is promising as a screening tool for low-resource settings because it is economical and provides immediate results. ASCO and WHO both recommend Visual inspection with acetic acid for basic settings i.e. developing and underdeveloped countries which lack the necessary resources of an organized cytology or HPV based screening programme[6,8]. When VIA based screening is initiated as population based screening it is important to ensure quality control and validated training and evaluation procedures.

In a study by Sankarnarayan et al, a single round of VIA screening by trained nurses led to increased detection of pre invasive disease and reduced mortality. (incidence hazard ratio 0·75 [95% CI 0·55–0·95] and mortality hazard ratio 0·65 [0·47–0·89])[75].

Poli UR et al, published a 7 years' experience using VIA in a community-based program in rural Andhra Pradesh, with a test positivity rates of 10.75% and prevalence of CIN2+ lesions as 1.05%. The rates of biopsy proven high-grade squamous intraepithelial lesions (HSILs) was 0.48% and low-grade squamous intraepithelial lesions (LSILs) was 0.28%[76].

Another study by Saleh HS et al, VIA had a higher sensitivity of 90% versus 50% with Pap smear. However, specificity of VIA was low at 37% compared to 93.5% with cytology[77].

In a recent meta-analysis of 32 studies the pooled estimates for VIA sensitivity and specificity were 0.69 (95% CI 0.54–0.81) and 0.87 (95% CI 0.79–0.92), respectively[78]. A recent review by Adsul et al of 20 studies including a total of 313,553 women at 12 different sites across India has been published[79]. They have reported that in 10 studies at cut off of CIN 2+, sensitivity for VIA ranged from 16.6% to 82.6%, and specificity 82.1% to 96.8%. At CIN 3+, the sensitivity ranged from 7.7% to 67.9%, and specificity from 87.4% to 96.7%. They concluded that to facilitate community based implementation of cervical cancer screening programs in India, there needs to be standardization of training to maintain competency of test providers; collaborations with community-based organizations for increased participation and use of 'screen and treat' method to reduce loss to follow-up[75]. VIA screening was associated with 31% reduction in cervical cancer mortality (RR = 0.69; 95% CI = 0.54 to 0.88; P = .003) and a better compliance in screened population than controls (86.3% versus 72.3%)[80].

WHO in a recent guidance has advocated that the lower age limit of cervical screening should not be under 30 years in LMICs and VIA permits a single-visit approach in low and middle-income countries (LMICs) in view of immediate test results and screen and treat approach[6]. In a modelling study for ascertaining the best screening strategy in developing countries, screening women once in their lifetime using VIA, at 35 years of age, reduced the lifetime risk of cancer by approximately 25 to 36 percent. The relative risk further reduced by an extra 40 percent with two rounds of screenings (at 35 and 40 years of age) [76,77].

To conclude, VIA is a feasible method to initiate mass screening in resource poor settings which lack an organized screening program. Once VIA based population screening has been implemented efforts should also be made to initiate HPV testing,

development and strengthening of infrastructure for diagnosis and treatment of preinvasive and invasive cancer.
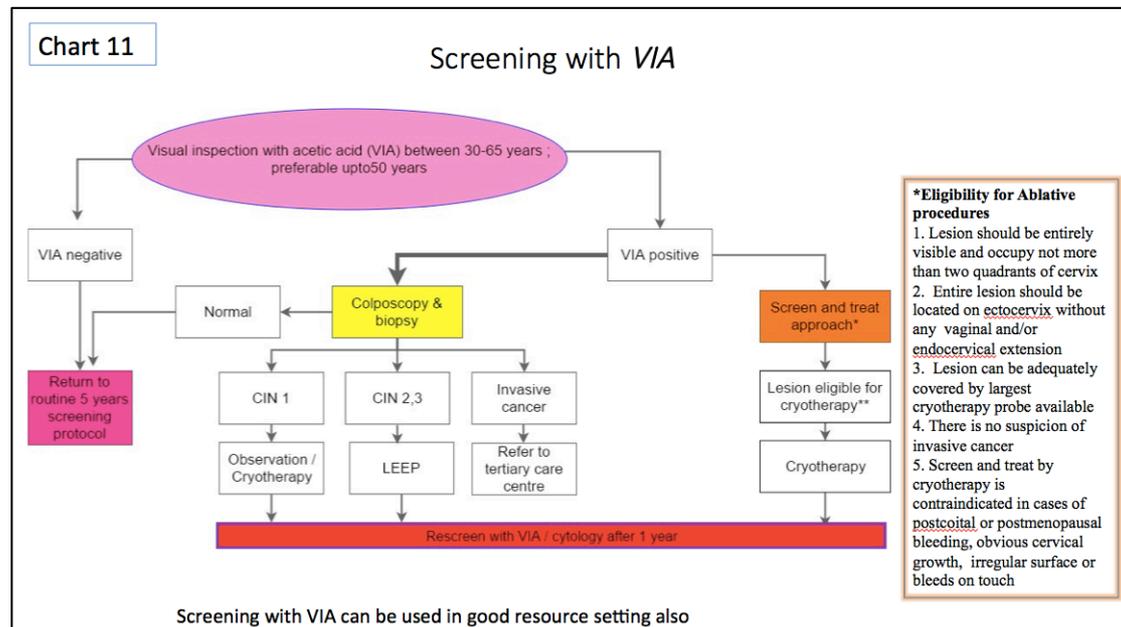


Fig 12 : Screening with VIA

**Recommendations for VIA as a screening tool: (Chart 11)**

1. VIA can be used as a screening tool between 30-65 years. ( level A)
2. In limited resource settings VIA should be done at least one to three times in a lifetime. (Level B)
3. Screening by VIA should not be continued beyond 50 years, especially in menopausal women as transformation zone recedes into the endocervical canal and prevents it from being fully visible. (Level A)

*Recommendations for VIA as a triage tool:*

1. In basic settings, visual assessment for treatment may be used after positive HPV DNA testing results. In cases of abnormal or positive triage results, treatment should be offered. When facility for colposcopy is available, it is preferable to do it before treatment. (Level B)

2. If VIA was used as primary screening, women with abnormal result should receive treatment. (Level C)

3. Women with negative triage results should receive follow-up in 12 months. (Level B)

## 4. Newer modalities

Infection with HPV leads to integration of viral DNA into the host DNA activating the proto-oncogenes to oncogenes, or deactivation of tumor suppressor genes; with consequent enhanced the rate of cell proliferation and cervical intraepithelial neoplasia (CIN)[81].

In young sexually active women, 90% of HPV infections are transient and only persistent infection predisposes to cervical neoplasia. HPV-DNA testing is highly sensitive test but cannot differentiate between transient and persistent infection.

The new generation of screening tests have evolved to identify markers that can discriminate between the transient and the persistent infections which if left untreated may progress to precancer and cancer. . These markers are promising as they increase the PPV and specificity of the test and at the same time also reduce colposcopy referrals and unnecessary interventions.

## p16 and Ki 67

The biomarker p16$^{INK4a}$, a cyclin dependent kinase inhibitor is present at very low levels in normal cells, while it is over-expressed in pre-cancer and cancer. Ki-67 is a nuclear protein that is expressed in proliferating cells and a higher Ki-67 index has been correlated with poor histopathological grades of disease[82]. For p16 immunostaining Gustinucci et al, showed sensitivity and specificity of 91% and 64% for CIN2+ respectively in ASCUS smears (n=213) and 77% and 64% for LSIL smears (n=186) respectively[82]. The sensitivity and specificity for p16/Ki-67has been variably reported between 64% to 98% and 43% to 81% across several studies for diagnosis of CIN 2+ disease in low grade smears and this difference is due to variation in sample size and interpretation of results[83,84,85].

Wang R et al showed that significantly high number of p16 +ve cases of CIN-1 progressed as compared to p16 negative cases (27% versus 7%) and p16 protein staining had a high negative predictive value of 93% for progression to CIN II-III[86].

In a study by Da Costa et al, ROC curves showed significant cutoff points of 0.396 and 0.026 for p16[INK4a] and Ki-67 ratios, respectively, as predictors of progression.

In a metaanalysis of 5 studies, higher agreement for a CIN2+ and CIN3+ diagnosis with H&E morphology in conjunction with p16[INK4a] was observed compared with H&E morphology alone[87].

The LAST guidelines concluded that p16 can be used to confirm a diagnosis of a high-grade lesion in CIN-2/ CIN1-2 on H/E morphology. p16 positive "CIN 2" will be labelled as "HSIL" while if p16 negative will be labelled "LSIL". p16 staining will decide further treatment and follow up in CIN 2 lesions and can be used to distinguish potential high-grade lesion from a benign mimic[88].

In a prospective analysis of the NTCC randomized controlled trial for HPV positive women, 8.8% [95% CI 5·8–11·8] p16 positive women had CIN2 or worse lesions compared to 3·7% [1·9–5·4] p16-negative women. Surveillance showed more CIN3 or worse was detected in more p16-positive women (4·4% [2·3–6·6]) than in p16-negative women (1·3% [0·2–2·3]; RR 3·90 [95% CI 1·57–9·68]). [89].

**DNA Methylation**

Changes in DNA methylation cause defective gene expression, genetic instability through faulty condensation of chromosomes, and silencing of mobile DNAs such as jumping genes (transposons) and viruses. Methylation can be measured as a simple reflex in HPV positive cases using quantitative methylation-specific PCR (QMSP)[90].

Methylation studies have been done on several genes of human genome namely MAL, CADMI, PAX-1, SOX-1 etc. A combination of *CADM1 and MAL in* hrHPV+ women from a screening population showed a sensitivity of 68% (95% CI 50– 81%) and a specificity of 75% (95% CI 70–80%) for CIN 2+ disease. At another cut-off , the sensitivity of *CADM1/ MAL* was 84% (95% CI 72–93%), the specificity was 52% (95% CI 48–57%), and the positive predictive value (PPV) was 25% (95% CI 17– 32%), with an AUC of 0.72 [91].

In a self-sampling study, DNA methylation testing for MAL and miR-124 was comparable with cytology as a triage tool in HPV positive women on self- collected specimens. The sensitivity for CIN2+ was 70.5% (95% CI 66.1–75.0%) and 70.8% (95% CI 66.1–75.4%) for methylation and cytology triage, respectively, while the

PPV was significantly lower (31.7%, 95% CI 26.3–37.1%, p < 0.001) for DNA methylation triage than for cytology triage (50.3%, 95% CI 42.3–58.4%)[92].

PAX1, SOX1and POU4F3 have been studies in Asian women showing promising results. The sensitivity of *PAX1* was 64% and for *SOX1* was 71%, while the specificity was 91 and 77%, respectively, and the AUC was 0.77 and 0.83, respectively. The sensitivity and specificity of *POU4F3* test taking CIN3+ cutoff is 74%, and 89%, respectively with an AUC of 0.86[93,94].

**E6 and E7 mRNA**

In a study in HR HPV positive women, E6/E7 mRNA positivity rate was 68.29 % in women tested once and 69.56 % in women tested twice in high grade lesions. The positivity rate was 89.28 to 84 % in women tested once and varied from 77.77 to 70 % when tested twice in women with low-grade lesions[95]. In a comparative study on HR HPV E6/E7 mRNA and HR-HPV DNA in cervical cancer screening, HR-HPV E6/E7 mRNA test showed a higher specificity than HPV DNA tests for high-grade lesions (61.4%, 54.3%, 55.7%, respectively, P < 0.05) and also a higher positive predictive value (75.9%, 74.8%, 74.6% respectively) with largest area of ROC curve and the best diagnostic value[96].

In another study, the sensitivity of HPV E6/E7 mRNA expression was lower than HPV DNA positive test , the HPV E6/E7 mRNA assay showed significantly higher specificity than the HPV DNA assay (88.6% vs. 48.1%) in normal cytology samples[97].

A large study on 396 samples demonstrated equal sensitivity for APTIMA E6E7 (AHPV) and Hybrid capture 2 for diagnosis of invasive cancer. The sensitivity of AHPV and HC2 were 95.2% and 94.9% respectively with an agreement of 97.2% (kappa 0.7; 95%CI: 0.54-0.87)[98].

**HPV self-sampling**

Across various studies the acceptability of HPV self-sampling is high due to its convenience, less discomfort and anxiety, ease of procedure and more convenience[99,100].

The sensitivity of self-collected samples are variable across literature, between 60 to 90%, with moderate to good agreement[101,102,103,104,105].

## 5. Screening in immunocompromised women

Infection with HIV increases a woman's lifetime risk of developing cervical premalignant and malignant lesions, approximately 10% HIV positive women develop CIN2+ each year compared to 1–2% among HIV negative women[106]. In a meta-analysis by Gary et al the prevalence of invasive cervical cancer was 4% and CIN2+ lesions was 8.5% among HIV positive women[107]. In a cross-sectional Indian study[106], 1,128 HIV-infected women underwent VIA, VILI, cytology, HPV testing, and colposcopy. Regarding diagnostic performance of various modalities in HIV infected population cytology was found to have s higher specificity than VIA and HPV testing but lower sensitivity than HPV testing. Sequential combinations of HPV testing and VIA; HPV testing and VIA; improved the specificity with minimal reduction in sensitivity .[107,108] VIA is a practical and useful screening test for HIV-infected women. The consideration for Prolonging the screening interval from 3 to 5 years for HIV infected screen-negative women, is applicable to HIV-negative women in current practice [109].

**FOGSI recommendations:**

1.Screening of HIV positive women should start in the first year of diagnosis irrespective of age. (Level A)

2. In order to enhance the coverage ART services should be integrated with cervical screening program. (Level B)

3. In good resource settings screening should be continued as per age recommendations every 3 yearly upto 65 years. (Level A)

4. In limited resource settings VIA should be done every 3 yearly upto 50 years. (Level B)

**6. Screening in pregnancy:** The available evidence is insufficient to currently recommend the guidelines for pregnant or postpartum women and therefore, the practice points are based on expert consensus.

**Recommendations:**

1. Pregnant women should undergo Speculum examination at first visit. If normal defer screening till postpartum period (6 weeks). (Level B)

2. If speculum examination suggestive of abnormality follow routine screening protocols. (Level B)

## 7. Management of Cervical intra epithelial neiplasia (CIN)

In women who have been diagnosed with CIN on biopsy which may or may not be colposcopy guided, further management is guided by the preceding cytology and HPV report if available. This is important in the case of low grade CIN (CIN1) where the preceding cytology report can indicate the future CIN3+ risk. A preceding low grade cytology (ASCUS-LSIL/HPV+) has a 5 year risk of 3.8% versus 15% with preceding high grade cytology[30].

Thus, in chart 12, follow up with preceding low grade cytology is follow up at 1 and 2 years, whereas with high grade smear more intensive follow up – cytology at 6months or even a diagnostic excision procedure is justified due to the discrepancy in cytology and histology reports. At any time during follow up if testing is postive, a repeat colposcopy and biopsy is recommended and subsequent management as per the histopathology report. CIN 1 is thought to be the histologic manifestation of HPV infection. CIN 1 is also associated with non-oncogenic HPV types, and among the oncogenic ones, HPV-16 is less common than in CIN 3[110]. The natural history of CIN 1 is similar to ASCUS/HPV + and LSIL in the presence of normal colposcopy and biopsy (see table with pap smear and risk of CIN), suggesting similar follow-up and management [111].

Thus, treatment for CIN 1 preceded by lesser abnormalities is only warranted when it persists for 2 years. Treatment can be in the form of ablative procedure if the criteria for ablation are fulfilled, or else excisional procedure is done.

CIN 1 preceded by high grade smear abnormalities can be kept on intensive follow-up (due to the higher risk of CIN 3) if colposcopy is adequate and complete and there is no endocervical involvement. If the converse situation, diagnostic excision is done. If high grade smear abnormality persists for 12 months and no lesion is seen on colposcopy also diagnostic excision is warranted.

Recommendations (Chart 12)

1. Women diagnosed with CIN 1on histology and preceded by low grade pap smear should be should be advised to continue with follow up under supervision (Level A).

2. In women with CIN 1 persisting for 2 years appropriate treatment should be advised (Level A).

3. Both ablation and excision are appropriate modalities for treatment. (Level B)

4. CIN 1 preceded by high grade smear abnormalities should be advised an intense follow-up or immediate treatment (Level B)

5. If high grade smear abnormality persists for 12 months and no lesion is seen on colposcopy a diagnostic excision is warranted. (Level B)

**Management of CIN in younger women (Chart 13 )**

Differences are – HPV is not part of follow up. CIN1 is a transient or stable HPV infection with minimal cancer risk, thus treatment is not recommended in the first instance. HPV testing does not form part of follow-up in women less than 30 years.

If CIN 1 preceded by lesser abnormalities persists for 1 year repeat colposcopy is done. If high grade abnormality persists for 1 year, diagnostic excision is done. This is based on the evidence from ASCCP that 5 year CIN3+ risk is 16-28% after ASC-H and HSIL in CIN1 but 5 year cancer risk is virtually nil. However, women with discrepancy in cytology and biopsy reports at the end of one year are subject to diagnostic excision to rule out underlying high-grade disease.

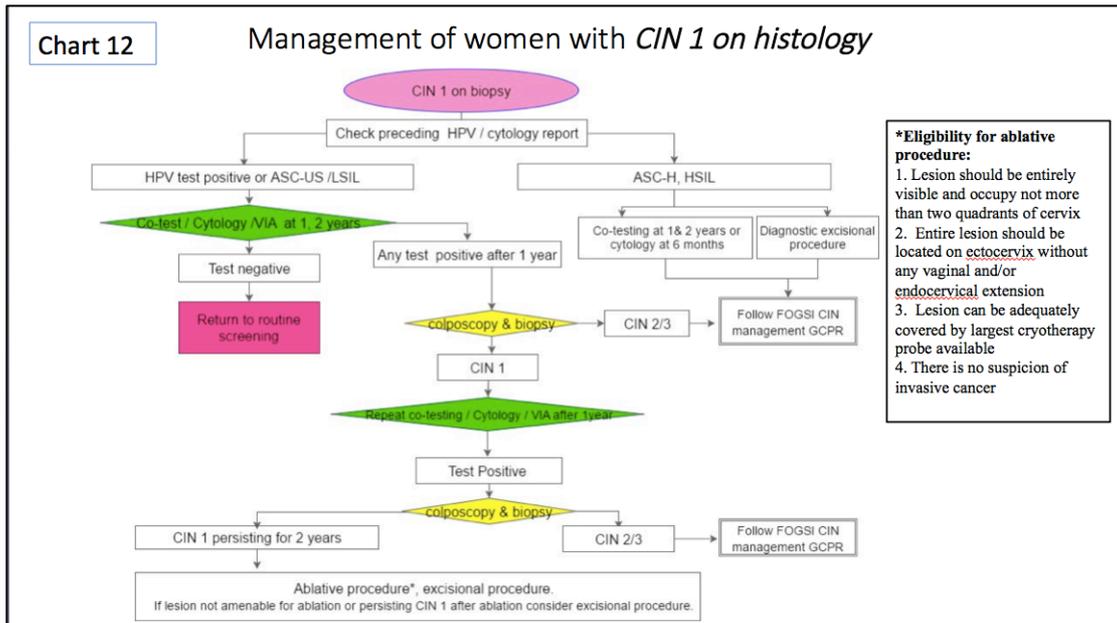Regression rates are high, especially in younger women, and progression to CIN 2+ is uncommon [112].

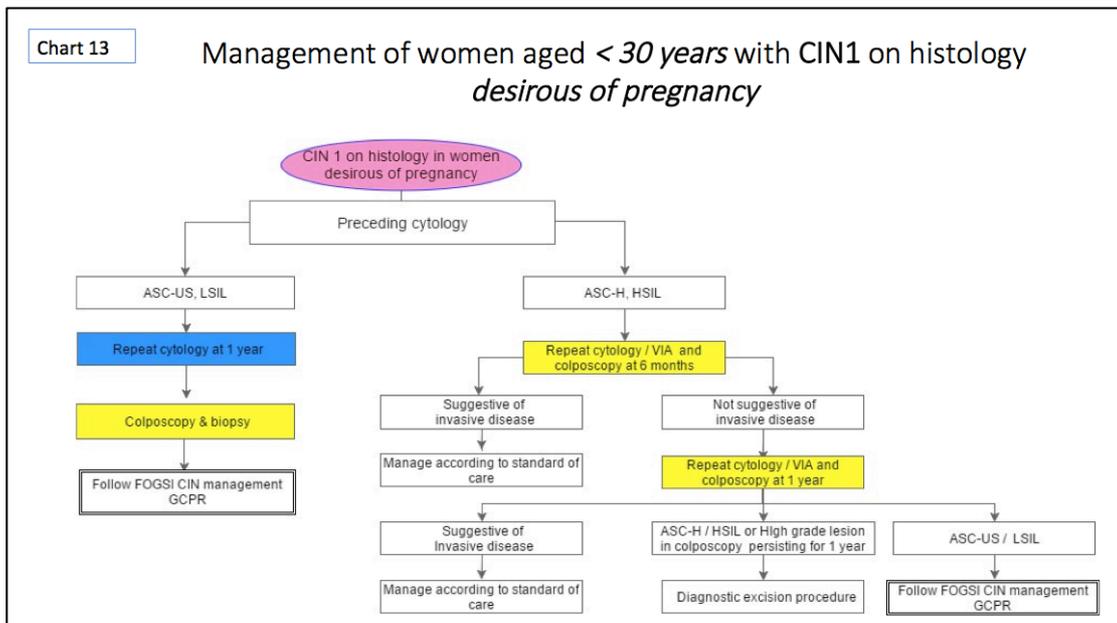**Fig 13: Management of women with *CIN 1 on histology***



 **Fig 14: Management of women aged *< 30 years* with CIN1 on histology *desirous of pregnancy***

**Recommendations (Chart12, 13)**

1. There is no role of HPV testing during follow-up and treatment (Level A)
2. If high grade abnormality persists for one year, treatment is acceptable (Level B)

## Cervical intraepithelial neoplasia (CIN) 2,3

As per WHO, 1-2% women are diagnosed to have CIN2+ every year[6]. Up to 50% of CIN 2+ can regress over time. Thus, treatment is recommended if CIN2+ persists for 2 years. For CIN3+ treatment is recommended, as risk of progression is high enough (approximately 30-50%). Thus, women with a diagnosis of CIN3 is harboring the immediate precursor to invasive cancer and should not be observed, irrespective of age or concern about future fertility.

Where CIN 2 or 3 is not individually specified, management algorithm followed is as per chart 14. With high grade CIN, if colposcopy is adequate, both excision and ablation are adequate modalities of treatment[113]. However, excision is preferred for large lesions or lesions not fully accessible for ablation. Diagnostic excisional procedure is recommended for recurrent CIN, if colposcopy is inadequate or if there is endocervical involvement, and ablation is unacceptable in these situations. Keeping CIN2/3 on follow up without treatment in not acceptable as the primary therapy, neither is immediate hysterectomy for the same.

If the margins of a diagnostic excision are positive, cytology and ECC can be repeated after 6 months. This is preferable to the alternative of doing a repeat diagnostic excision, which is acceptable in individual cases. If a repeat excision is not technically feasible, hysterectomy is acceptable as well.
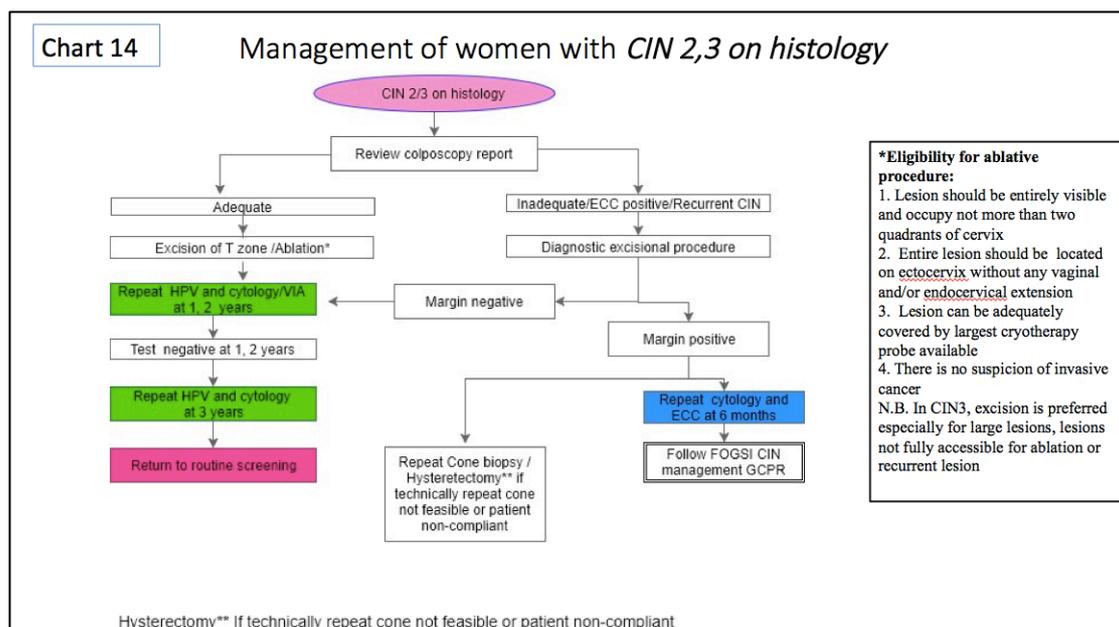


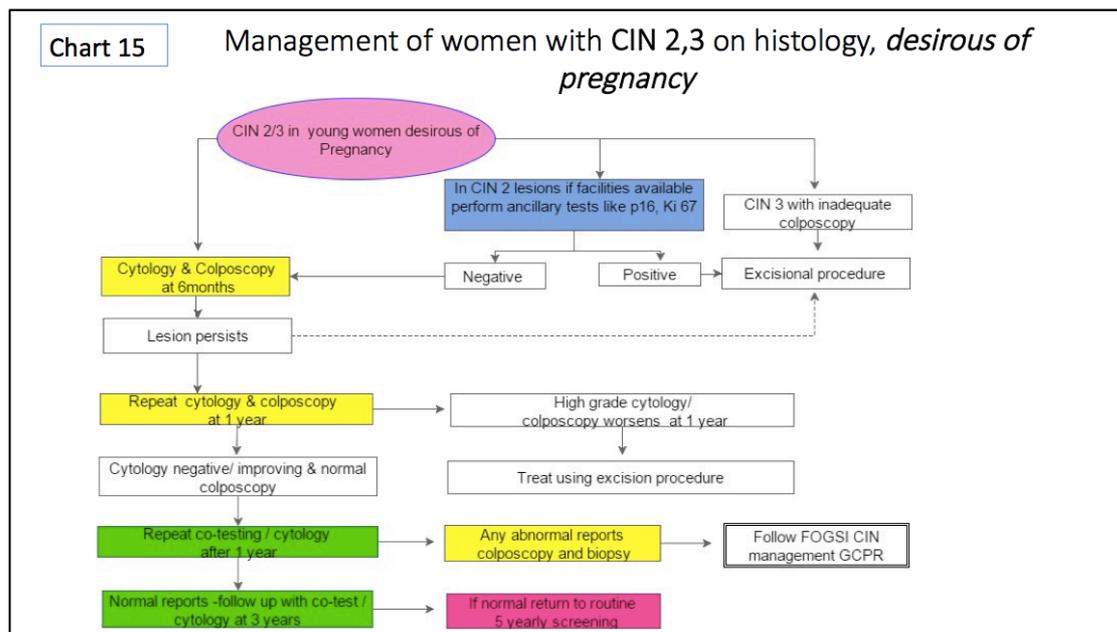**Fig 15 : Management of women with _CIN 2,3 on histology_**

**Fig 16 : Management of women with CIN 2,3 on histology,** *desirous of pregnancy*

**Recommendations (Chart 14)**

1. With high grade CIN, if colposcopy is adequate, both excision and ablation are adequate modalities of treatment (Level A)

2. Excision is recommended if recurrent CIN, endocervical involvement or colposcopy is inadequate. (Level A)

3. Follow-up of CIN2/3 without treatment is unacceptable (Level B)

4. Immediate hysterectomy for CIN2/3 is unacceptable (Level B)

5. For positive margins after excision, cytology and ECC can be repeated after 6 months (Level A)

6. Hysterectomy as an alternative to repeat excision is feasible (Level C)

In the context of young women, the aim is to treat women with high risk of developing invasive disease. Those who are not at high risk are observed and kept on follow-up as treatment can have adverse consequences on future pregnancy. The aim is also to protect them from harms of over-treatment[114].

As per chart 15, where CIN 2/3 is diagnosed (not individually specified), observation and follow up at 6 months and 1 year with cytology and colposcopy is an acceptable alternative to treatment, provided colposcopy is adequate. After two consecutive

negative cytology results, an additional co-test at 1 year and 3 years is recommended. If either is abnormal, colposcopy and biopsy is performed and managed accordingly.

If CIN 2 is specified, triage biomarkers may be done if available. Observation is preferred to treatment, unless triage biomarkers are positive. Treatment is an acceptable alternative if biomarkers are negative. During observation, if the cytology becomes high grade or the colposcopic appearance of the lesion worsens at 1 year, repeat biopsy is recommended[114,115].

If CIN3 is specified and colposcopy is inadequate, treatment (excisional procedure) is recommended. In CIN2 with positive biomarkers, or if CIN 2,3 persists during follow up at 6months & 1 year, treatment is recommended.

**Recommendations (chart 15)**

1.  Observation without treatment for unspecified CIN 2/3 is acceptable provided colposcopy is adequate (Level A)
2.  If CIN2/3 persists during follow up at 6month and 1 year, treatment is recommended (Level A)
3.  Observation is preferred to treatment if CIN2 is specified (Level A)
4.  Treatment is preferred to observation if CIN3 is specified or colposcopy is inadequate (Level A)
5.  Excision is preferred to ablation (Level B)

**CIN in pregnancy -** Treatment for CIN during pregnancy is deferred unless invasive cancer is suspected. Patient is kept on follow up with colposcopy and cytology not more frequently than every 12 weeks. Repeat biopsy is recommended only if invasive cancer is suspected. Post-partum evaluation is deferred for 6 weeks postpartum[114].
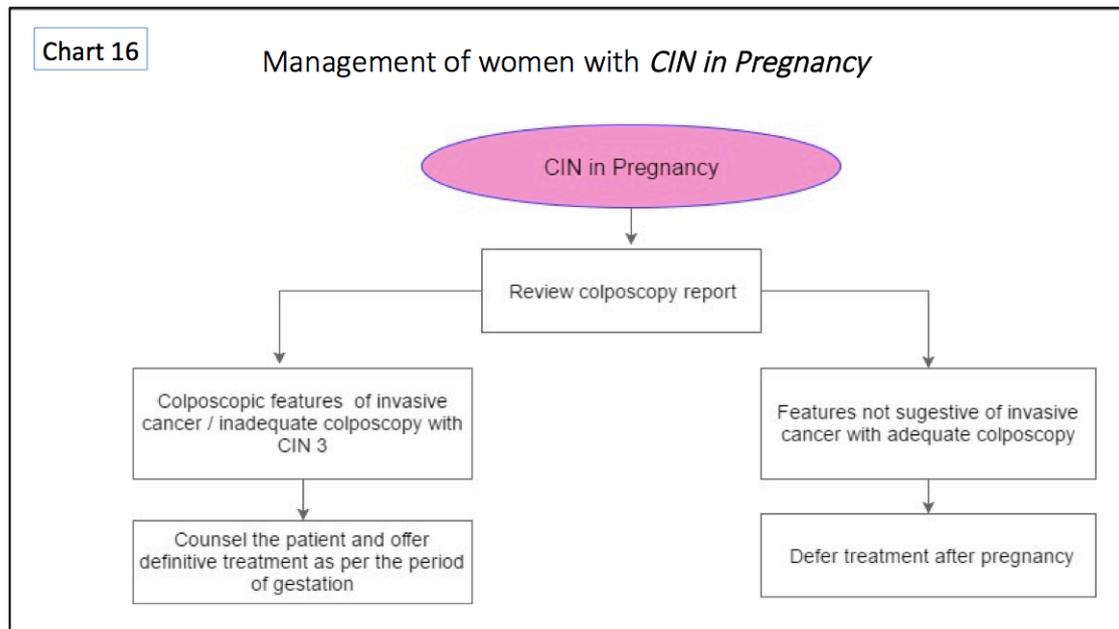
**Fig 17 : Management of women with CIN in pregnancy**

**Recommendations (Chart 16, Fig 17)**

1. Diagnostic excision is advisable only if invasive cancer is suspected (Level A)
2. Defer post-partum evaluation to 6 weeks (Level A)


**Organization of Outreach Community Camp for Cervical Cancer Screening**

In India there is poor access to screening and treatment services. Following barriers are identified for access to screening services

- Lack of knowledge about cervical cancer

- No perceived need by women hence no apparent reason for screening

- Geographic and economic inaccessibility of services

- Discomfort during internal examination

- Lack of support from families and communities

- Stigma: Fear of getting screened positive

- Lack of knowledge about availability of treatment options among the population

- Preventive health still of primitive importance/value

- Female issues still a neglected component of the society

- Mismatch between the demand & the supply.

Cancer cervix can be prevented by population based screening, which aims to detect the disease at a precancerous stage when it is amenable to simple treatment and cure. Outreach camps can be helpful in overcoming above mentioned barriers. The main purpose of organizing cervical cancer screening camps in outreach settings is to increase access to quality screening services for women of lower socio-economic category living in hard to reach areas and to offer immediate treatment if required thus by reducing the loss to follow-up. Conducting outreach camps among group of women helps to create awareness, moreover women get motivated by seeing others getting screened and feel relaxed.

For a successful execution of outreach community camps following strategies can be planned-

SITE SELECTION CRITERIA: An initial visit should be done to the selected site to ensure that following criteria's are fulfilled-

- Approachable to the community
- Clean
- Adequate running water supply/ backup
- Functional toilet
- Well ventilated and airy
- Well lighted
- Electricity/Backup

- Room for processing of instruments-cleaning and High-Level Disinfectants (HLD)

- Screening room with space of at least two examination tables

- Room/Space for counselling (to maintain privacy)

- Space/room for registration

- Waiting area with sitting arrangement for clients

- Sitting arrangement for accompanying person

HUMAN RESOURCE: In successful execution of outreach camps, role of following are crucial-

*Demand Generation-*

- Community field workers- They are the main interface with the community who can mobilize the clients. USHA (Urban Social Health Activist Urban Social Health Activist), ASHA (Accredited Social Health Activist), AWW (*Anganwadi* workers) or NGO (Non-government organization) workers of selected site can support in demand generation and spreading information about proposed camp among community

- Local heads in the community like MLC (Member of Legislative Council), religious leaders, or any other influential person may be of help in orienting the community.

*Service Provision-*

- Trained doctor- for providing screening services

- Paramedical staff- for documentation work, counselling, supporting doctor during screening and ensuring follow-up of VIA screened positive women

- Support staff- for cleaning/sterilisation of equipment and managing group during camp

**DO'S AND DON'TS FOR PLANNING OF CAMP**

| *Do's –* | *Don'ts –* |
|---|---|
| • Plan the camp well in advance. <br><br> • Assign roles and responsibilities clearly. <br><br> • Convey the VIA positive result to the client in a non-threatening and empathetic way. <br><br> • Direct and follow up with the VIA positive client needing services at higher centres. <br><br> • Motivate clients to go back in the community and encourage others also, for screening. <br><br> • Ensure correct data is captured on the formats. | • Don't overload the camp with clients. In order to assure quality of service delivery one doctor can screen up to 50 clients in a day |

**STRATEGY FOR DEMAND GENERATION:** Women may not always be motivated to attend screening just because a new service is being provided. Carefully designed targeted messages and strategies can be used to encourage women to take advantage of new services. Cervical cancer prevention services typically focus on women who are older (aged 30 to 59 years) and in need of other health services. Women's participation in screening can be improved by seeking out eligible women when they are reached for the health services, to attend to other problems. They can be provided

other health check-ups like Hb, BP monitoring, Blood sugar screening and nutritional counseling etc. simultaneously with cervical cancer screening if feasible. Information brochures, leaflets, pamphlets, flyers, posters and banners can be used as awareness generation and propagation tool. These screening camps can also be organized for-school/college staff, widow's homes, handicraft workers, physically challenged women, women inmates in prison and workplace settings etc.

## MANAGEMENT OF CLIENTS DURING THE CAMP

- Assign roles and responsibilities and prepare a flow chart for women's movement during a screening camp

- Ensure better services for the clients on the camp day.

- Organizing team must ensure the smooth functioning.

- Provision of logistic for clients, sitting arrangements and drinking water

- Provision of logistic sitting arrangements, food/ snacks for the staff

- Separate designated areas for registration and counselling must be ensured.

- Confidentiality of the screening results and other details are to be ensured.

## COUNSELLING AND FOLLOW-UP

Counselling is an important component of service delivery. It satisfies women's information needs, reduces their anxiety and fear, and explores possible barriers to treatment, follow-up. Following counselling related procedures should be followed as appropriate.

- Individual or group pre-screening counseling

- Counseling about screening results

- Individual counseling for women who need further care

- Pre/post treatment counseling for women treated with cryotherapy

**DO'S AND DON'TS FOR COUNSELLING:**

*Do's –*

- Make eye contact, listen attentively and take note of her body language (posture, facial expression, eye contact).

- Try to understand her feelings and point of view.

- Use open-ended questions to invite more than "yes" or "no" answers.

- Be empathetic: place yourself in the woman's situation.

- Use simple language and terms that the woman understands.

- Answer her questions truthfully and allow enough time for the session.

- To cross check -ask the woman is she pregnant. If she is pregnant, ask her the age of gestation and explain her accordingly.

- After completing the pelvic examination, ask the woman if she is more comfortable discussing the results while lying on or sitting on the table.

- Inform the women about VIA test findings

- Give detailed information about the treatment option

- Describe the benefits and effectiveness of cryotherapy

- Explain how the cryotherapy procedure is done and the potential side effects, also ensure that the woman understands

- Encourage the woman to ask questions and discuss her condition

-  it is better to take consent for treatment along with screening consent, avoid 2 separate consents)Give the woman some time to decide if she has doubts, invite her to return later to inform you of what she (and possibly her family) has decided

- If you noted something for which you wish to refer her to a higher level for further examination or tests: Explain why, where and when she must go, and whom to see.

- Stress the importance of keeping this appointment.

- Invite her to return if she has any questions or concerns about this appointment, and respond or find answers from someone who knows.

*Don'ts –*

- Appear to be distracted (looking at your watch, answering the phone).

- Use of harsh tone of voice, or act impatient.

- Allow interruptions during the visit.

- Interrupt the woman.

- Be critical, judgmental or rude.

- Overwhelm the woman with too much detail or irrelevant information.

- Use medical words the woman does not understand.

- Insensitively share the results without asking readiness of the client to hear the results

- Force a decision regarding treatment rather ask consent of the client for treatment.

- Let the client leave without understanding the treatment options she has in her hands

- Be in rush and not let the client ask questions she may have in her mind

**ENSURING QUALITY SERVICES:** Good quality of services must be ensured through training of service providers on standard protocols. A monitoring checklist is given in

table below will help in assessing the logistic arrangements, record filling and maintenance and infection prevention practices during the outreach community camp.

| Sl.no | MONITORING CHECKLIST |
|-------|---------------------|
| A | *INSTRUMENTS & MATERIALS REQUIRED FOR VIA & CRYOTHERAPY* |
| 1. | Drum -autoclaved (with speculum, sponge holding forceps, steel bowl, green towel) |
| 2. | Drum -autoclaved containing sterile cotton balls |
| 3. | Acetic acid |
| 4. | Container for preparing acetic acid solution |
| 5. | Steal bowl |
| 6. | Distilled water |
| 7. | Measuring Cylinder, syringe- 5ml |
| 8. | Sodium hypochlorite |
| 9. | 70% ethanol spray |
| 10. | Normal Saline |
| 11. | Cusco Speculum |
| 12. | Sponge Holder |
| 13. | Mackintosch |
| 14. | Mouth mask |
| 15. | Gloves |
| 16. | Towel |
| 17. | Dustbins 3 |
| 18. | Disposable bags |
| 19. | Gloves boxes 3, plastic gloves (if required) |

| | |
|---|---|
| 20. | Boiler with plug |
| 21. | Writing pad/ board with clip for holding the forms |
| 22. | Address cards, prescription pad |
| 23. | Table/Trolley |
| 24. | Table fan |
| 25. | Trolley, green towel |
| 26. | Cryo machine with probe & Filled N2O Cylinder *(optional)* |
| 27. | Extension board (for boiling instruments) |
| 28. | Torch - 2, extra cells(LED torch preferably with four batteries) |
| 29. | Register for keeping log |
| 30. | Gynaec examination table, stool, writing table, chairs, Hand sanitizer/Soap etc |
| B | *RECORD FILLING AND MAINTENANCE* |
| 31. | Consent forms to be signed by the beneficiaries |
| 32. | Client Record  Sheet |
| 33. | Report Card/Follow up instructions card. |
| 34. | Storage of client records properly done |
| C | *INFECTION PREVENTION* |
| 35. | Autoclaved instruments |
| 36. | Hand hygiene practices followed by all Staff |
| 37. | Mackintosh placed on the examination table |
| 38. | Gloves are changed after examining every patient |
| 39. | Aprons are worn by the Provider |
| 40. | Hand rub/ Hand sanitizer available and used |
| 41. | Mouth masks are used while Procedure |

| 42. | Examination table cleaned in-between screening of clients |
|---|---|
| | Bleaching solution prepared in adequate amount & used instruments dipped in it for 10 minutes. |

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<center>**Section 2: HPV Vaccination**</center>

## Introduction

Persistent infection with oncogenic types of human papillomavirus has been shown to be the necessary cause of cervical cancer.[1]

About 6.6% of Indian women in the general population have HPV infection at any given time.[2] Most of the time, HPV infection does not cause any manifestations and is self-limited. It has been observed that approximately 70% of all newly acquired HPV infections are cleared within 1 year, and approximately another 20% in next one year. In some women infection persists and may cause cervical cancer and other manifestations including oropharyngeal cancers, anogenital cancers and genital warts. Human Papillomavirus is a non-enveloped, small, double stranded DNA of the family papilloma viridae. The viral genome consists of 7200–8000 base pairs and the early fragments of the genome regulates DNA replication (E1, E2), transcription (E2) and cell transformation (E5, E6, E7) and late fragments (L1&L2) encode structural proteins of the virion. Purified L1 protein form empty shells resembling a virus, called virus-like particles (VLPs).

HPV are largely non-lytic and restricted to the epithelium. Antibodies will be formed in serum against many different viral proteins. The most characterized antibodies formed are those directed against conformational epitopes of the L1 capsid and detectable antibodies are not found in all infected persons. In a study by Carter et al, 54%–69% of women who were found to be infected with HPV 6, 16, or 18 infections had type-specific antibody.[3]

More than 100 types of HPV have been recognized on the basis of DNA sequence showing genomic differences.[2] Infection with HPV types 1 and 2 cause warts in some individuals while

HPV-5 may cause lifelong infection without any clinical manifestation. Infection with HPV types 6 and 11 can cause laryngeal papillomatosis and genital warts. HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73, and 82 are considered to be carcinogenic.[4] HPV 16 and 18 are high risk types and accounts for over 75% of invasive cervical cancers in South Asia.[5]

In India approximately 80% of cervical cancer and 63% of high grade cervical lesions are linked with infection due HPV 16 and 18.[6] Age and sexual activity are the major

influencing factors for HPV transmission. About 75% of sexually active individuals harbor at least one

HPV type.[7] Risk of HPV infection can be reduced by an HPV type-specific targeted vaccine. Abstinence or lifetime mutual monogamy is helpful in preventing genital HPV infection. There is limited evidence to suggest protective role of condoms and male circumcision against HPV infection.[8]

## Types of HPV Vaccines

In India two HPV vaccines are available presently which are globally licensed: quadrivalent vaccine (Gardasil™, Merck Inc.) and bivalent (Cervarix™, Glaxo Smith Kline, Ltd.).[9] The third, a nonavalent vaccine, was US FDA approved in 2014, but is not yet available in India. Both vaccine comprises of type-specific major capsid protein L1 synthesised by recombinant DNA technology. The vaccines do not lead to any infection in the individual.

Characteristics of HPV vaccine:

|  | Bivalent Cervarix | Quadrivalent Gardasil | Nonavalent Gardasil 9 |
|---|---|---|---|
| Manufacturers | Glaxo Smith Kline | Merck | Merck |
| HPV types in vaccine | 16,18 | 6,11,16,18 | 6,11,16,18,31,33,45,52,58 |
| Adjuvant in vaccine | AS04:500ug aluminium hydroxide 50ug 3-O-desacyl-4'-monophosphoryl lipid A | AAHS:225ug amorphous aluminium hydroxyphosphate sulfate | AAHS:500ug amorphous aluminium hydroxyphosphate sulfate |

## Safety:

Extensive data on the safety of HPV vaccines are available from clinical trials and population programs. Globally, more than 270 million doses have been administered with no serious adverse event linked to the HPV vaccine and with an excellent safety profile. Adverse events reported after HPV vaccine administration were generally mild in intensity and like those expected after any vaccination. These included vaccination site pain, tenderness, swelling, fever, headache, myalgia and gastrointestinal symptoms.[10,11] The safety of HPV vaccines is

monitored by the WHO Global advisory committee for vaccine safety (GACVS) which regularly reviews the evidence related to their safety. GACVS in 2016 stated that decreased use of safe and effective vaccines based on weak evidence can lead to harm and the available evidence does not raise any concern related to the safe use of HPV vaccines.[12]

**Contraindications:**

- Allergy to yeast and yeast products

- Allergy to previous dose of HPV vaccine (irrespective of brand)

- Pregnancy and lactation

- Patients with moderate or severe acute illnesses

**Recommendations**

1. HPV vaccine is safe to use and all available safety data of HPV vaccines do not suggest any safety concern. (Level A)

**Efficacy of HPV vaccine**

Prior to licensing, quadrivalent vaccine was evaluated in 2 large Phase III trials (in addition to a previous Phase II study) which are named as FUTURE I [13] and FUTURE II[14]. These studies involved more than 150 sites in 16 countries and involved more than 17,000 participants.

In Future 1 trial 5455 women between 16 to 24 years were enrolled, and the efficacy of the

qHPV vaccine in terms of protection provided against CIN 2/3 and AIS caused by HPV16 or HPV18 was evaluated over a follow up period of 3 years. In this trial the females who were naive to HPV16/18 one month after 3rd dose, the protection against combined endpoint was found to be 100% (95% CI 94-100%).[13]

In Future II Study Group 12,167 women aged 15-26 years were included and the efficacy against CIN 2/3 and AIS caused by HPV16/18 was found to be 98% (95% CI, 86-100%) 3 years after the first dose.[15]

A combined analysis of two phase III studies, reported by the Future II study group in 2007 included 17,622 females aged 15-26 years, who at the baseline were found to be infected with one or more oncogenic vaccine related HPV type. The effectiveness of quadrivalent vaccine was 100% (95% CI 79-100%) against CIN 2/3 and AIS after 3 doses and 3 years of observation period in women who were seronegative at enrolment.[16]

Clinical trials of bHPV vaccine have been conducted in more than 18,000 women worldwide receiving three dose schedule (at 0, 1& 6 months). The Patricia trial was conducted in women aged 15-25 years, it was a randomized double blind controlled trial. They were vaccinated with HPV16/18 AS04 adjuvant vaccine at 0, 1&6months. The mean follow up was 34.9months after the 3rddose and the efficacy of vaccine was 92.9% against CIN2+ associated with HPV 16/18 in the primary analysis and in the according to protocol cohort for efficacy it was 98.1%.[17] Another trial the Costa Rica vaccine trial was conducted on 7466 healthy females aged 18-25 years and were randomized into HPV arm (HPV16/18 vaccine) and control arm (Hepatitis A vaccine). A median follow up of 4+ years showed the vaccine efficacy of 89.8%

against HPV 16/18 associated CIN2+ and about 59.9% with oncogenic HPV other than 16/18. [18]

On a 15-month follow-up of 1113 women aged 15-25 years vaccinated with bivalent HPV 16/18 vaccine, 90% efficacy was found against HPV16/18-related CIN-2/3 and AIS. Study included women who were HPV DNA negative at baseline for vaccine type and received one dose of the vaccine in modified intention to treat analysis. Over a of 4–5 years follow up in the participants did not showed waning immunity.[19]

In 2014 the USFDA approved 9-valent HPV vaccine (9vHPV). This vaccine targets HPV types 6, 11, 16, and 18, as well as HPV types 31, 33, 45, 52, and 58. These five additional HPV types account for about 15% of cancers (14% for females; 4% for males) and approximately 25% of ≥CIN2.[82] Dosing, precautions and contraindications for HPV 9 are similar to that of Quadrivalent vaccine.[20] At present 9-valent vaccine is not available in India.

**Duration of protection:**

Women has risk of getting HPV infection anytime during her sexual life, therefore the overall effectiveness of vaccine depends on duration of protection period. The studies have shown heterogeneity in the methods used for assessing immunogenicity, histopathological and virological end points and statistical power issues, so it is difficult to interpret the long term effectiveness of the vaccine. Association of higher antibody levels with longer duration of protection is questionable. Vaccine efficacy depends on its ability in disease prevention for a long period of time. To- date the follow-up of bHPV vaccine is 9.4 years and for qHPV vaccine is 8 years. The follow up of vaccination upto 9 years has shown that vaccine is immunogenic and well tolerated.[21-34]

**Target Population, Dosage Schedule and Duration of Protection**

Centre for Disease Control and Prevention (CDC) and the American College of Obstetricians and Gynecologists (ACOG)[35] recommends routine HPV (bHPV or qHPV)vaccination for girls and boys of 11-12 years. According to the Indian Academy of Paediatrics Committee on Immunisation (IAPCOI) all females should receive HPV vaccine, if affordable starting from age 9 years and catch up vaccination at age 13 through 45 years if not already vaccinated.[36]

The ideal time for vaccination would be before start of sexual activity, as vaccine is most effective before getting exposed to HPV infection.[37] Girls age less than 9 should not receive HPV vaccines. The geometric mean titres (GMTs) induced by both vaccines are 2 times higher in girls aged 10-15 than older girls aged 16 to 26[38-40]. Catch-up vaccination is recommended up to 26 years of age for girls if they have not taken the vaccination at target age.

In older age groups(15-26years) the intention-to-treat naive cohort (in women who were HPV DNA negative at baseline, were HPV seronegative for HPV 6/11/16/18 and had normal cytology) who received at least 1 dose of the qHPV vaccine, 100% efficacy was seen in preventing HPV-type related CIN 3 disease.[38]

**Cross-protection against non-vaccine HPV types:**

Quadrivalent HPV vaccine provides cross protection to some extent against other hrHPV types apart from HPV 16&18. The efficacy of qHPV vaccine against CIN2+ caused by any of the 10 most common oncogenic non-vaccine types was found to be 32.5% (95% CI: 6.0–51.9) in the FUTURE I and FUTURE II studies[41].

A systematic review and meta-analysis included 46 publications, and most of them had a maximum follow-up period of four years. During this follow-up a borderline protective effect with a pooled RR of 0.80 (95% CI: 0.62-1.02) was observed in all CIN2+ with catch-up vaccination. There was reduction in VIN2+ and VaIN2+

lesions, and condyloma with catch-up vaccination. The long-term effect of this catch up vaccination in prevention of cervical cancer and its related mortality is still unclear.[42] Another study by Munoz N in 2009 concluded that the qHPV vaccine is effective when given to women aged 24-45 years, not found to be infected with relevant HPV types at enrolment.[43]

**Recommendation**s

2.  HPV vaccines are licensed for use in females aged 9-45 years; however the preferred     target age group is 9-14 years. (Level A)
3.  Vaccination in sexually active females it may be less effective, but may provide some benefit as exposure to all vaccine types previously is unlikely. (Level B)
4.  Females aged 15-25 years should be considered for catch-up vaccination programme only if resources are available.(Level B)

**Dosage Schedules:**
HPV vaccines are administered as a 2-dose series for those who initiate vaccination at ages 9 through 14 years, and a 3-dose series for those who initiate at ages 15 through 26 years, and for immunocompromised persons. The number of recommended doses is based on age at administration of the first dose. The minimum interval of 6 months is recommended between 2 dose schedules for girls aged < 15 years. An additional third dose is required at least 6 months after the first dose if the interval between two doses is less than 5 month[20,44] Though the

maximum interval between the doses is not recommended, but a duration of 12–15 months complete the vaccination schedule and before sexual activity.
A large multicentric cohort study was conducted in 188 clusters(9 location) in India,in which 17,729 unmarried girls aged 10-18 years were vaccinated with QHPV vaccine. There were divided into four cohorts- those who received 3 doses at 0,2 &6 months, 2 doses at 0 &6 months, 2 doses at 0&2months by default and those who received one dose by default. It was found that the geometric mean avidity indices were non-inferior when the fewer than 3 doses by design and default were compared to 3 doses

of vaccine. Those girls who received fewer than 3 doses by design and default had detectable neutralizing antibody concentration to all the 4 HPV vaccine types but the concentrations were much lower in those who received one dose.[45]

**Recommendations:**

5.  Girls aged 9-14 years of age should receive 2 doses of HPV vaccine at least 6 months apart, although the interval between 2 doses can be extended to 12-15 months in circumstances where second dose is not repeated within 6 months. (Level A)

6.  Catch up vaccination can be offered to females more than 15 years till 26 years. They should receive 3 doses, however the second dose should be given after 1or 2 month (depending on the vaccine that is used) and third dose after 6 month of the first dose. (Level A)

7.  Older Girls/ women who have been sexually active should be counselled regarding reduced efficacy of HPV vaccine and the importance of screening from age 25 years. (Level A)

**Special Situations**

*Interrupted Dosage Schedule*

In case vaccine schedule is interrupted or there is delay in taking second or third dose, the vaccine series need not be restarted.[20,44]

Dose interruption may also occur due to non-compliance to scheduled vaccination or pregnancy. For dose interruption due to pregnancy, please refer to appropriate section entitled 'Vaccination in pregnancy'.

In a study conducted in Vietnam the 3 doses of qHPV vaccine given as a standard dosing schedule (0, 2, 6 months) was compared with 3 alternative dosing schedules (0,3,9 months; 0, 6,12 months; or 0, 12, 24 months)[46]. The use of 2 alternative dosing schedules (at 0, 3, 9 months and at 0, 6, 12 months)

was non inferior in terms of antibody concentrations when compared with a standard dosing schedule. However the group that received doses at 0, 12, 24 months did not met the non-inferiority criteria.[47] Follow-up of the women enrolled in this study did not show inferior immune responses after >2.5 years of follow-up after the third dose.[47]

**Recommendation:**

8.   A 2-dose schedule with a minimum interval of 6 months between 2 doses is recommended for girls aged <15 years. If the time period between two vaccine doses is less than 5 months, a third dose is recommended 6 months after the first dose.(Level A)

9.   There is no recommendation for maximum interval between two doses, but a period not more than 12–15 months is required to complete vaccination schedule. (Level B)

10.   There is no need to restart vaccine schedule, when second or third dose is delayed. (Level A)

**Co-administration with Other Vaccines**

Bivalent or quadrivalent vaccines can be given with other age-specific vaccines, such as diphtheria, acellular pertussis and tetanus alone or in combination (dTpa, dTpa-IPV vaccines) and also with quadrivalent meningococcal conjugate vaccines. Giving age specific vaccines with a separate syringe at different sites in the same visit increases the chance of receiving vaccine on schedule and will reduce the number of visits to the health care center.[20]

Concomitant use of HPV vaccine with recombinant Hepatitis B vaccine is found to be safe in girls and women ages 16 to 23 years. The adverse reactions were similar in concomitant vaccination when compared to Gardasil or recombinant Hepatitis B alone [48,49] There is no clinically relevant interference reported with antibody response to any of the components of these vaccines. Immunogenicity and safety is proven for co-administration with combined Hepatitis A and B vaccine.[50] Studies have shown that the vaccines are generally well tolerated, and the immune responses were non inferior when used in combination compared with non-concomitant administration [51-53]. The immunogenicity of dTpa-IPV and the first dose of HPV vaccine was found to be similar.

**Recommendation**

11.    HPV vaccine can be safely administered with other age appropriate vaccines. (Level A)

*Victims of Sexual Abuse*

The risk of HPV infection increases with sexual abuse and assault and is attributable to abuse itself, increased risk for future victimization, and subsequent engagement in at-risk behavior. Though, vaccination does not provide any benefit in terms of viral clearance or protection against disease progression in an already acquired infection, but it provides protection against the infection which are not yet acquired. Children who undergo sexual abuse have more chance of engaging in unsafe sexual practices at an

earlier age. Health care workers should be aware of the need for HPV vaccination in females with history of sexual abuse or assault.[54]
Advisory Committee on Immunization Practices (ACIP) and Centre for Disease Control (CDC) recommends HPV vaccination for both male and female sexual assault survivors aged 9–21 years and 9–26 years respectively. In case of sexual assault 3 dose schedule is followed with the first dose given

at time of initial examination and follow up doses, at first or 2nd month and second at 6th month after the first dose. American Academy of Paediatrics (AAP) also recommends HPV vaccination beginning at 9 years of age in sexually abused females. There are not enough studies to prove the efficacy of HPV vaccines in sexually abused or assaulted women.

**Recommendations**

12.    Sexual assault survivors should be given 3 dose course of HPV vaccine, with first dose at the time of initial examination. (Level B)

*Administration in Older Age Groups*

The vaccine is most effective in sexually naive girls and young female i.e, before they are exposed to HPV infection. The new infections usually do not progress to CIN 2 or worse and are associated with a low risk of developing cervical cancer in older women. So vaccine prophylaxis in order to prevent infections with high risk HPV types leading to cancer at older ages is questionable.[55-57]

Although HPV incidence decreases with age, women more than 25 years remain at risk of developing new HPV infections [43,58]. In the study by Castellsague X et al which evaluated the efficacy of Quadrivalent vaccine in women aged 24-45, it was found that the efficacy of vaccine against the combined incidence of persistent infection, CIN or extra genital lesion related to HPV6/11/16/18 in the per-protocol efficacy(PPE) population was 88.7% (95% CI: 78.1, 94.8), and the efficacy in seropositive and DNA negative women for HPV 6/11/16/18 vaccine type at the time of enrolment was 66.9%(95% CI: 4.3, 90.6). Due to limitations of the study, the efficacy against CIN 2/3 was not statistically significant in women aged 24–45 years.[59] Antibody levels are also found to be lower in older age group but remain substantially higher than those induced

by natural infection for at least 6 years following the first dose. To conclude the utility of Quadrivalent HPV vaccine in adult women is still not adequately known but there could be small benefit on individual basis by vaccinating adult women. In United States for women older than 26 years HPV vaccines are not licensed currently [60].

However, strong HPV-16 and 18-specific antibody response was reported in women up to 55 years in a study, but there was decrease in the antibody titers which was age-dependent. Antibody titers in 46- 55yrs age group was 8-fold higher than titers due to natural infection in 15- 25year age group. Evidence for efficacy in age > 45 years are lacking.[61]

**Recommendations:**

13. For women aged 25 to 45, the first priority should be given to cervical cancer screening. Cervical cancer screening and HPV vaccination are not mutually exclusive. (Level A)

14. When women aged 25-45 years are vaccinated, they should be counselled about reduced efficacy of the vaccine. (Level A)

*Women with abnormal Pap smear or positive HPV test or Previous HPV Lesions*

Women with cervical screening abnormalities may have associated one or more types of HPV infection. As the abnormality in Pap's increases, the chance of infection with HPV 16 or 18 increases with a decrease in expected benefit of vaccine.

Although these women can be vaccinated still, but with prior proper counselling that vaccine will not help in treating the already existing HPV infection, HPV associated pre-cancerous lesion, cancer or anogenital warts[62].

**Recommendations**

15.   Women with abnormal Pap/ HPV screening results or previous HPV lesions can be vaccinated if they desire, however they should be counselled about the lack of any therapeutic effects on the existing pathology and reduced efficacy of the vaccine in older women. (Level B)

*Special Populations*

*Vaccination- Pregnancy and lactation*

HPV vaccination should not be initiated during pregnancy, but if a woman becomes pregnant after starting the vaccination schedule, termination is not required and the remaining dose(s) should be given after delivery without repeating the initial dose(s).

If a vaccine dose is given during pregnancy, no intervention is needed and lactation is not a contraindication for HPV vaccination.

The data available from various studies did not show any increase in the risk of adverse effects either in mother or baby due to HPV vaccination given in lactation period.[20,63] Although clinical trials that led to licensure of both the vaccines excluded pregnant women, and the study participants had to avoid conception within 2 months of the last dose. However among HPV vaccine recipients (who participated in clinical trials of both vaccines), more than 6500 pregnancies have occurred[64]. There was no significant differences observed in rates of any specific pregnancy outcomes between the vaccine and the placebo recipients. The manufacturers of both vaccine companies maintained registry of the women who were inadvertently vaccinated during pregnancy and they did not find any increase in the rate of major congenital malformation and spontaneous abortions.[63]

**Recommendations**

16.   HPV vaccine should not be initiated during pregnancy. (Level A) However if pregnancy is detected after initiation of vaccine it is

advisable to discontinue the schedule and continue the vaccine 6 weeks post pregnancy. (Level A)

*Immunocompromised Women*

HIV infection:

HPV and HIV infection share a common route of transmission, and therefore, the vulnerable populations often coincide. In HIV positive women HPV infection is significantly more common as compared to HIV negative women across all age groups. HPV infection increases with age in HIV positive women.[65] HPV Vaccine was well

tolerated in HIV-infected children aged 7–12 years and was found to be safe and immunogenic. The occurrence of adverse events was similar in qHPV and placebo recipients and were infrequent, except for a more frequent injection site reactions in Quadrivalent HPV recipients.[66] Quadrivalent vaccine was found to be both safe and highly immunogenic in HIV-1 infected men & women with ≥ 95% seroconversion for each of the HPV types.[67]

**Recommendation**:

17.    HIV positive girls should be advised to start HPV vaccination from 9-14 years and should be prescribed 3 dose schedule (0,1,6 months) (Level B)

*Vaccination of Males*

The quadrivalent HPV vaccine for boys and young men was licensed by the US Advisory Committee in Immunization Practices (ACIP) in 2009, 3 years after licensing qHPV for girls [68]. It is upto 90% effective in young men in prevention of  HPV infection which includes genital warts and anal epithelial neoplasia.[69,70]It also provides protection against high-risk HPV(HPV16&18), which are associated with various other cancers.[71]

Quadrivalent vaccine is recommended for boys aged 11–21 years, and has been approved up to age 26, for prevention of genital warts and anal cancer.[72] In MSM (Men Who Have Sex with Men) also, it is approved upto the age of 26.[72] Quadrivalent Vaccination programs will help in reducing the burden of HPV-related diseases and cancers[73-76].

There has been a significant increase in the vaccination coverage among boys in USA, in 2013, approximately 34.6% of boys received one dose of qHPV vaccine in age group 13–17 years.[74] Though there has been an increase in the vaccination coverage, only 13.9% of boys completed the recommended three-dose of vaccine.[77]

Australia first initiated a government-funded universal HPV vaccination for school boys in February 2013.[78] Routine qHPV vaccination is recommended for both boys and girls by US ACIP, for the children aged 9–18 years who are uninsured or under insured and the HPV vaccination is financed by the Vaccines for Children (VFC) Program.[72,79] Those who were not eligible for VFC vaccination were electively covered by state.[80]

In 2008 national immunization program was started in UK which provided universal coverage for girls only,[81] similar to most of the European countries.[82,83] In 2014 Austria was first to initiate gender neutral HPV vaccination program.[84] In Canada 11 provinces and territories immunized only girls in HPV vaccination program.

In countries where vaccination in males is introduced they had already achieved optimal vaccination coverage in girls. Vaccination in girls still remain the priority and vaccination in males in developing countries and in resource limited countries are not in practice.

**Recommendations:**

18. Vaccination in males is not recommended at present in Indian setting. (Level C)

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