About OMICS Group

OMICS Group International is an amalgamation of Open Access publications and worldwide international science conferences and events. Established in the year 2007 with the sole aim of making the information on Sciences and technology ‘Open Access’, OMICS Group publishes 400 online open access scholarly journals in all aspects of Science, Engineering, Management and Technology journals. OMICS Group has been instrumental in taking the knowledge on Science & technology to the doorsteps of ordinary men and women. Research Scholars, Students, Libraries, Educational Institutions, Research centers and the industry are main stakeholders that benefitted greatly from this knowledge dissemination. OMICS Group also organizes 300 International conferences annually across the globe, where knowledge transfer takes place through debates, round table discussions, poster presentations, workshops, symposia and exhibitions.
About OMICS Group Conferences

OMICS Group International is a pioneer and leading science event organizer, which publishes around 400 open access journals and conducts over 300 Medical, Clinical, Engineering, Life Sciences, Pharma scientific conferences all over the globe annually with the support of more than 1000 scientific associations and 30,000 editorial board members and 3.5 million followers to its credit.

OMICS Group has organized 500 conferences, workshops and national symposiums across the major cities including San Francisco, Las Vegas, San Antonio, Omaha, Orlando, Raleigh, Santa Clara, Chicago, Philadelphia, Baltimore, United Kingdom, Valencia, Dubai, Beijing, Hyderabad, Bengaluru and Mumbai.
ADVANCES IN GYNAECOLOGIC CYTOLOGY

Role of immunomarkers in increasing diagnostic accuracy of lesions of cervix

Dr Nandini N, Manoli (Prof, Patho),
Dr Nandish S Manoli (Prof, OBG),
Dr Shweta Kulkarni (PG, Patho),
Dr A.P Chandrashekar (Prof & HOD OBG),
Dr Anjali Siddesh (Prof, OBG)
JSS Medical College, a constituent of JSS University, Mysore, Karnataka, India
INTRODUCTION:

- Cancer remains one of the world’s leading causes of death and a major health and economic burden.

- Worldwide, cervical cancer is the third most commonly diagnosed cancer in women (approx 530000 new cases) resulting in 275000 deaths annually.

World wide cancer incidence globe scan 2008
INTRODUCTION:

- Since the **pap test** was introduced in the 1940s, there has been an approximately 70% reduction in the incidence of squamous cell cervical cancers in many developed countries by the application of organized and opportunistic screening programmes.

- **70% reduction** in the incidence of squamous cell cervical cancers in many developed countries by the application of organized and opportunistic screening programmes.

- The efficacy of the pap test, however, is hampered by **high interobserver variability and high false negative and false positive rates**
INTRODUCTION:

- Investigators have attempted by various means to enhance the sensitivity of the pap test.

- First, by the introduction of liquid based methods to address issues of specimen collection and preparation and later,

- use of computer assisted screening systems to address the screening errors and to improve the screening efficiency and disease detection.
INTRODUCTION:

- High risk HPV DNA testing has a very high sensitivity for the detection of high grade cervical disease,
- It has a very low specificity and positive predictive value.
- The use of biomarkers has demonstrated the ability to overcome the issues with both false positive and false negative results
- leading to improved positive predictive value of cervical screening results.

Signal amplification methods - HC2 test
INTRODUCTION:

• Numerous protein bio-markers for the detection of cervical disease have been identified.

• Many of these proteins are involved in cell cycle regulation, signal transduction, DNA replication and cellular proliferation.

A simplified diagram illustrating cell-cycle functions of candidate biomarkers of cervical neoplasia

Peter Baldwin, Ronald Laskey & Nicholas Coleman
INTRODUCTION:

• Biomarkers currently under investigation for use in cervical cancer screening
• That appear to improve the detection of women at greatest risk for developing cervical cancer, include Ki-67, P16^{INK4A}, BD ProEx c and HPVL1

• These biomarkers are reported
• to have a role in the triage of indeterminate cytology cases,
• discrimination of true high grade cervical dysplasia from mimics in histology
• serve as predictive markers to identify lesions most likely to progress to high grade cervical disease and cancer.
The protein P-16\textsuperscript{INK4A} derived from the host P-16\textsuperscript{INK4A} /CDKN2A tumor suppressor gene, the protein has been identified as a biomarker for transforming HPV infection and therefore can be used as a surrogate marker of HR-HPV infection.

The protein accumulates in the nucleus and cytoplasm of affected cells and can be detected by immunocytochemistry.
P-16 INK4A

• Several studies have tested it in either LBC or cell block preparations and the majority have demonstrated the effectiveness of P-16 INK4A for improving the cytological detection of HSIL.

• These studies showed that P-16 INK4A has good specificity (SP) and positive predictive value (PPV).

REF: p16INK4a immunocytochemistry on cell blocks as an adjunct to cervical cytology: Potential reflex testing on specially prepared cell blocks from residual liquid-based cytology specimens
MIB-1 (Ki-67)

• Ki-67 is an antigen that identifies proliferating cells and is expressed in all phases of the cell cycle. MIB-1 is a monoclonal antibody that detects this antigen in the nuclei of fixed cells or tissues embedded in paraffin.

• When HPV infection leads to increased epithelial cell proliferation in infected tissues, increased Ki-67 staining can be an indicator of HPV infection.

SCC was the diagnosis on cell block finding. Strong and diffuse staining was observed for Ki-67 (B). Original magnification ×400 (A, B). HE staining (A); DAB staining (B).

ref   p16(INK4A) and Ki-67 immunostaining on cell blocks from residual ThinPrep material is helpful in identifying significant preneoplastic cervical lesions Huiqiong Bao, Yilin Wu
MIB-1 (Ki-67)

- In dysplasia and carcinoma, Ki-67 expression extends above the basal one third of the epithelium and the thickness of the epithelium and the number of positive cells increases.

- There is a significant positive correlation between ascending grade of squamous intraepithelial lesion and labelling index.
BD-Pro Ex C

• BD-pro ex C is a protein based biomarker reagent containing antibodies to the nuclear proteins minichromosome maintenance protein 2 (MCM2) and topoisomerase II alpha (TOP2A), proteins that have been shown to accumulate in HPV transformed cells.

• They are both over expressed when the S phase cell cycle induction is aberrant.

• Advantages of these are exclusively nuclear biomarkers which are easier to detect than those producing cytoplasmic staining.
Biomarker expression in low-grade squamous intraepithelial lesions and high-grade squamous intraepithelial lesions detected in liquid-based cytology samples and Cytoactiv HPV L1 staining performed on conventional Pap smears lesions in cervical cytology specimens. Ki-67, p16^INK4a, BD ProEx C, and BD SurePath Plus

Ref Charlotte A. Brown, et al Role of Protein Biomarkers in the Detection of High-Grade Disease in Cervical Cancer Screening Programs Journal of Oncology Volume 2012 (2012),
L1 capsid protein

- L1 is primarily the name of the major capsid protein of HPVs.

- L1 is also the name of an antibody against a protein of the HPV16 capsid that is expressed in the early productive phase of the viral life cycle and is progressively lost during cervical carcinogenesis.

REF  Ralf Hilfrich
The combination of L1 and P-16 INK4A antibodies in LBC samples and cell blocks has been proposed for prognostic prediction of LSIL.
E-Cadherin and β-catenin

- The disruption of intercellular adhesions is an important component of the acquisition of invasive properties in epithelial malignancies.

- Alterations in the cell-cell adhesion complex E-cadherin/β-catenin, have been implicated in the oncogenesis of carcinomas arising from various anatomic sites and have been correlated with adverse clinico pathological parameters.
E-Cadherin and β-catenin

- Impairment of E-cadherin and β-catenin expression is very frequent in early stage cervical cancers.

- Reduced expression of E-cadherin is significantly associated with overall survival and disease free survival in the patients with cervical carcinoma.

- It serves as an indicator of aggressive clinical behaviour and could suggest the use of adjuvant therapy in early stages of the disease.

E-cadherin in CIN III of Cervix

D. Mocuta, D. Craiut, T. Pop, Elena Lazar

Recent Advances:

- Chromosome studies show genetic changes in chromosome 3, which include changes in 3p loss and 3q gain.

- 3q gain
  A gain at the segment between chromosome band 3q24/25 and band 3q28 is associated with HSIL.

- These observations have led to the hypothesis that this genetic aberration might play a pivotal role in the transition from pre-invasive lesions to invasive cervical cancer.

- 3q abnormalities
  Human telomerase RNA gene (hTERC) and PIK3CA gene are located in Chromosome segment 3q26.
Figure 1. Cells positive and negative for 3q26 gain.

http://www.plosone.org/article/info:doi/10.1371/journal.pone.0039101
Table showing the different categories of cervical lesions and markers

<table>
<thead>
<tr>
<th>Categories of lesions</th>
<th>Number of lesions(60)</th>
<th>P-16</th>
<th>Ki-67</th>
<th>Ecadherin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Squamous cell carcinoma</td>
<td>2</td>
<td>++</td>
<td>++</td>
<td>_</td>
</tr>
<tr>
<td>Dysplasia</td>
<td>21</td>
<td>+</td>
<td>+</td>
<td>_</td>
</tr>
<tr>
<td>Inflammatory conditions</td>
<td>20</td>
<td>_</td>
<td>_</td>
<td>+</td>
</tr>
<tr>
<td>Normal smears(NILM)</td>
<td>17</td>
<td>_</td>
<td>_</td>
<td>++</td>
</tr>
</tbody>
</table>
Recent advances:

- p63 is located at 3q28.

- 3q26 gain identifies subset of LSILs with more aggressive biologic behaviour.

- Management of 3q positive will be colposcopy. 3q negative will be follow-up.

- p63/p73 and piK3CA are newer markers studied

Pathway showing relationship between p53, p63 and p73 and their role in tumorigenesis

Ref MP DeYoung and LW Ellisen
p63 and p73 in human cancer: defining the network
Oncogene (2007) 26, 5169–5183
Markers tested in combinations

Several studies have tested more than one biomarker on the same sample.

The complementary role of P16 and MIB-1 on LBC and Cell Block preparations which in combination is compared to LBC, improves diagnostic accuracy for HSIL and squamous cell carcinoma.

Conclusion:

• There are different methods for early detection of cervical cancer. They are
  
  • A) Biomarkers on tissue,
  • B) Serum levels of various human markers,
  • C) HPV testing
  • D) gene profiling.
Conclusion:

- The biomarkers or immunomarkers can be studied on various cytological specimens like
  - a) LBC,
  - b) Cell block
  - C) Histopathological biopsies

- To increase the sensitivity, specificity and diagnostic accuracy of cervical cancer
Conclusion:

- These marker studies are cost effective as compared to HPV testing which is more accurate.

- Thus improvement in diagnostic accuracy and cost effectiveness makes it useful to include biomarkers in single or in combinations in cervical cancer screening programme.

- These along with clinical colposcopy, VIA, conventional pap smear screening, LBC, cell block and histopathological biopsies will help to decrease the deaths by cervical cancer in developing countries.
References


• Nieh S F, Chen TY Chu, HC Lai and E Fu. Expression of P-16 INK4A in papanicoloau smears containing atypical squamous cells of undetermined significance from the uterine cervix. Gynaecologic oncology 91, p-201-208,2003
References


• Ralf Hilfrich. HPV L1 Detection as a Prognostic Marker for Management of HPV High Risk Positive Abnormal Pap Smears.


Acknowledgment

- Department of Pathology H.O.D, Faculty, Postgraduates and Technicians. JSS Medical College, Mysore.

- Department of Pathology, Kidwai Institute of Oncology, Bangalore.

- Department of OBG, JSS Medical College, Mysore.
THANK YOU

JSS MEDICAL COLLEGE
**Let Us Meet Again**

We welcome you all to our future conferences of OMICS Group International

Please Visit:

- [www.omicsgroup.com](http://www.omicsgroup.com)
- [www.conferenceseries.com](http://www.conferenceseries.com)
- [http://pathology.conferenceseries.com/](http://pathology.conferenceseries.com/)