Viruses that infect cutaneous and mucosal epithelial tissues, more than 95% of cervical cancers are positive for HPV DNA and a subset of HPVs, known as high risk types, mainly HPV 16 and HPV 18. These two types are mainly associated with cervical cancer (CaCx) development[1]. The viral genome of HPV 16 contains 7,904 bp and the HPV genome is composed of six early (E1, E2, E4, E5, E6, and E7) ORFs, two late (L1 and L2) ORFs, and a non-coding long control region (LCR). HPV 16 infects mucosal and cutaneous epithelia and form lesions that can persist and progress to cervical cancer[2]. In HPV-infected basal cells, every early transcript encoding E6, E7, E1, E2, E4, and E5 are expressed with their specific functions. For example, viral E1 and E2 proteins are expressed to maintain the viral DNA as an episome and they also help to facilitate the correct segregation of genomes during cell division[3]. In these early ORFs HPV 16 encodes three oncoproteins E6, E7 and E5. The role of two oncoproteins E6 and E7 in the formation of cervical cancer (CaCx) has been extensively studied. E7 protein inactivates pRB to cause continuous division of infected host cells and E6 induces the degradation of p53 to prevent apoptosis in HPV infected cells. But the role of another oncoprotein E5 in cervical carcinogenesis is not clearly understood. This article will review my current understanding of the role of HPV 16 E5 oncoprotein and its interacting partners in the progression of cervical cancer (CaCx)[4].

Papillomaviruses are a large group of viruses that have been found almost 20 different species in mammals. There are 100 different types of HPV already have been identified because of their medical importance[1].

LIFE CYCLE OF HPV

HPV is a circular dsDNA virus with a genome of 7,904 bp. The genome is divided into three regions where six early (E) ORFs and two late (L) ORFs are expressed. The viral genome carries all ORFs on a single sense strand and carries one upstream regulatory region (URR) or non-coding region (Figure 1)[5]. The virus shell of HPV is composed of two coat proteins L1 and L2, L1 is the primary structural element where L2 is the minor virion component and present at the center of pentavalent capsomers[1]. During genomic amplification, the expression of L1 and L2 is upregulated both at
There are five stages present in HPV life cycle, they are (a) Genome maintenance (occurs at basal layer); (b) Genome maintenance and cell proliferation (occurs at suprabasal layers); (c) Genome amplification (occurs at suprabasal layers); (d) Virus assembly and release (occurs at cornified layers); (e) Virus release (occurs at granular layers).

There are five stages present in HPV life cycle, they are (a) Genome maintenance (occurs at basal layer); (b) Genome maintenance and cell proliferation (occurs at suprabasal layers); (c) Genome amplification (occurs at suprabasal layers); (d) Virus assembly and release (occurs at cornified layers); (e) Virus release (occurs at cornified layers).

## ONCOPROTEINS OF HPV

HPV 16 four oncopogenic proteins: E6, E7, E5 and E4. The E6 and E7 oncoproteins are required for the inactivation of p53 and pRb tumor suppressors and to induce telomerase. The main function of E6 protein is to degrade p53, E6 forms a complex with p53 and the 100 kDa E6AP (ubiquitin ligase). This interaction results in the ubiquitination of p53 through the proteosomal pathway. The E7 protein inactivates and induces the degradation of the retinoblastoma protein (pRB), a cell cycle regulatory protein required for the G1/S transition and DNA synthesis. Besides modulating cell cycle control and inactivation of pRb, E7 protein has been implicated to downregulate the level of human toll-like receptors (hTLR) and the ER protein TAP-1. E6 and E7 both interact with many other cellular factors inducing tumor progression, immune evasion and genomic instability. The oncopenic activities of E6 and E7 are studied extensively and are well characterized. But the role of another oncoprotein E5 is not very clear to researchers. So, now we discuss about E5 oncogene with its special features, main functions and the role of CaCx development.

### E5: THE SMALLEST ONCOGENE OF HPV

HPV E5 proteins are less well characterized and they are localized mostly to the Golgi apparatus, endoplasmic reticulum and they also present in the nuclear membrane. HPV E5 proteins are hydrophobic in nature and contain 83 amino acids; it is the smallest oncogene of HPV. E5 exhibits transforming activity and induces a malignant phenotype to cooperate with E6 and E7 oncoprotein. E5 is also able to stimulate several cellular pathways through the activation of epidermal growth factor receptor (EGFR) and can upregulated vascular endothelial growth factor (VEGF). E5 has a primary role in proliferative and productive phase of viral life cycle. When the HPV genome is episomal, the E5-mRNA is the most abundant viral transcript and it plays an important role in the early phases of HPV-mediated transformation.

### Table 1 HPV proteins and their functions

<table>
<thead>
<tr>
<th>HPV Proteins</th>
<th>Molecular Weights</th>
<th>Functions</th>
</tr>
</thead>
<tbody>
<tr>
<td>L1</td>
<td>55 kda</td>
<td>Major viral capsid protein self-assembly in capsomers and capsids interacting with L2, Interacting with cell receptor</td>
</tr>
<tr>
<td>L2</td>
<td>70 kda</td>
<td>Minor viral capsid protein Interacting with DNA, Facilitating virion assembly Interacting with cell receptor</td>
</tr>
<tr>
<td>E1</td>
<td></td>
<td>Viral genome replication DNA helicase and ATPase activity Binding to specific DNA elements in the viral origin of replication and assembly into hexameric helicases with the assist of a second viral protein, E2.</td>
</tr>
<tr>
<td>E2</td>
<td>42 kda</td>
<td>Site specific DNA binding protein. Viral genome replication. Viral genome expression regulation Viral genome segregation during cell division Interacting with and recruits E1 to the origin. Playing a role in regulating viral transcription from the early promoter.</td>
</tr>
<tr>
<td>E4</td>
<td></td>
<td>Facilitating virus assembly and release Binding and collapsing the cytokeratin network. Preparing the progression of cells into mitosis by arresting them in the G2 phase of the cell cycle. Binding to mitochondria. Inducing the detachment mitochondria from microtubules. Induction of apoptosis.</td>
</tr>
<tr>
<td>E1^E4</td>
<td>10 kda</td>
<td>83 residues Cellular transformation and initiation of neoplasia. Being able to active epidermal growth factor receptor (EGFR) and other protein kinases. Inhibiting apoptosis and interacting with gap junction proteins.</td>
</tr>
<tr>
<td>E6</td>
<td>150 amino acids</td>
<td>Immortalization and transformation of infected epithelial cells. Disrupting normal cell growth and proliferation by binding to protein p53. Inducing telomerase and preventing cell differentiation.</td>
</tr>
<tr>
<td>E7</td>
<td>100 amino acids</td>
<td>Immortalization and transformation of infected epithelial cells. Disrupting normal cell growth and proliferation by binding to protein pRb.</td>
</tr>
</tbody>
</table>

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tumorogenesis[13]. There are multiple cellular effects found, which are associated to the expression of HPV16 E5. E5 binds the 16 K proteolipid subunit of the membrane proton pump and this binding may play an essential role in EGFRII over activation[19]. Some reports said that the binding region of E5 has been present to amino acids 54-78 or amino acids 41-54[20]. The HPV E5 ORF itself has been classified into four groups alpha, beta, gamma and delta which have oncogenic potential with different clinical manifestations. The E5 alpha protein is encoded by high risk HPV, whereas, the E5 gamma and delta proteins are encoded by low risk HPV’s. The expression and amino acid sequences of HPV 16 E5 were detected by Chang et al. In cervical intraepithelial neoplasia (CIN), low-grade squamous intraepithelial lesions (LSILs) and high-grade SILs. They reported that there are three anchors like α-helices present in amino acid sequence of E5 at residues 8-30, 37-52 and 58-76.

INTERACTION OF E5 WITH OTHER PROTEINS

Interaction with MAP Kinases

E5 protein of HPV 16 upregulates the signal cascade initiated by EGFR through mitogen activated protein kinases, which are serine/ threonine protein kinases in nature and are involved in directing cellular responses to a diverse array of stimuli such as osmotic stress, heat-shock, mitogens and this MAP kinase pathway is associated with both cell proliferation and differentiation[19]. Activated MAP kinase is translocated into the nucleus upon activation, where it can potentially phosphorylate transcription factors, such as c-Jun, c-Myc, and c-Tal1[20]. Many oncogenes such as ras, raf, src and mos transform cells by prolonging the activated stage of MAPKK. Some reports have described E5 gene is often deleted from the viral genome during viral DNA integration in the more advanced cervical neoplastic lesions, similarly several reports have described E5 does participate in transformation process when the HPV DNA is episcopal and it has a potential to be act as an oncogene which can show its transforming activity by downregulating the EGFR and increase the number of EGFR-Rs at cell surface[17]. Several reports observed that HPV 16 oncogene products could alter cellular MAP kinase activity and E5 gene was able to induce an increase in the MAP kinase activity. Some assays are help to determine the effect of E5 on MAP kinase signaling pathway, both E5 and mutant E5 are showing their activity in this assay conditions. The mutant E5 used this assay was increased the translation efficiency of the corresponding E5 mRNA by introducing Kozak sequence around the initiation ATG of the E5 gene. The result of this in vitro transcription-translation assay showed that mutant E5 was at least five times more efficiently translated in-vitro, but both wild type and mutant E5 demonstrated a similar increase in the MAP kinase activity[17].

Interaction with Calpactin I

Calpain I is a heterotetrameric protein complex that consists of two annexin A2 or lipidocin II subunits and two p11 subunits[18]. Annexin A2 is a 36 kD calcium binding proteins, associates with phospholipids in a calcium dependent manner and regulates different cellular processes such as membrane fusion and endocytosis[19]. Several reports observed that HPV 16 E5 binds calpain I and induces perinuclear membrane fusion events. E5 is present in vacuole membranes, and its C terminus is required for vacuole formation. Krawczyk et al 2011 reported that E5 associates with calpain I after transfection in COS cells, they done immunoprecipitation where they observed a 36 kDa polypeptide that coimmunoprecipitates with E5 was excised from a Coomassie blue-stained sodium dodecyl sulfate-polyacrylamide gel, digested with trypsin, and microsequenced. After this experiment they confirmed that there was a strong association present between E5 and annexin A2, E5 and p11 and annexin A2 and p11 in COS cells. They also observed that E5 promotes perinuclear membrane fusion with calpain I, to confirm this hypothesis they used HEC stable cell line which express HPV 16 E6 and it infected with retrovirus encoding E5. After 3 days they pulse-labeled the cell line for 30 min with Alexa Fluor 594 (red)-conjugated CTB and six hours later the same cells were pulse labeled with an Alexa Fluor 488 (green)-conjugate of CTB. Then they performed fluorescence colocalization and they found a small number of cells exhibited perinuclear vacuoles that showed two distinct populations of membrane vesicles had undergone fusion. It is already demonstrated that E5 induces a perinuclear redistribution of calpain I and the binding interaction with E5 and perinuclear localization are mediated by the C terminus of E5, which is known to be exposed to the cytoplasm[20]. In this study Krawczyk et al2011 identified calpain I as a new cellular target for the HPV 16 E5 oncoprotein and E5 have high affinity for calpain I.

Interaction with ER stress pathway proteins

HPV 16 E5 protein has the ability to bind several cellular proteins. The presence of viral proteins may activate cellular defense mechanisms and in particular the ER stress response. HPV 16E5 can suppress three main proteins of the ER stress pathway and they are cyclooxygenase-2 (COX-2), XBP-1 and IRE1a. It can alter splicing of XBP-1 which is a part of ER stress response pathway. In 2010 Sudarshan et al reported that high risk HPV 16 E5 protein has an effect on XBP-1 splicing. To study gene expression they used microarray approach and tried to capture alterations in the ER stress pathway. At first they constructed 16 E5 and E5 mutant in pLXN vector and then used EcoRI and BamH1 to clone the construct into the vector for stable expression. On the other hand they prepared human foreskin keratinocytes (HKFs) and human ectocervical cells (HECs) and treated them with chemical inducers of ER stress. During microarray each E5 cell lines was run against the corresponding pLXN cell lines. Next they performed reverse-transcriptase PCR (RT-PCR) and prepared cDNA by denaturing the RNA for 3 minutes at 80°C with oligod (T) and random hexamers. cDNA samples were diluted and used in real time RT-PCR. After real time RT-PCR, they quantified the ratio of spliced to unspliced XBP-1 transcripts in HKFs and HECs cell lines and also found E5 protein reduce the expression of XBP-1 in ER stress pathway. Then they looked to the mutant E5 and studied its role in these cell lines, in high risk HPVs the region of E5 protein where the mutations were made, were highly conserved. So, all mutant E5 proteins also showed reduction of XBP-1 expression. After all these experiments they reported that in ER stress pathway 179 genes were significantly altered by HPV 16 E5 protein[21].

In 2009 Kim et al reported that COX-2 expression is downregulated in cervical cancer lines C33A and SiHa. They concluded that HPV 16 E5 is able to lower COX-2 expression in cells co-expressing E6/E7, during viral replication[22]. However, other studies suggested that COX-2 protein levels did not correlate with the disease severity of HPV-induced cervical lesions or with HPV-positivity in primary and metastatic cervical cancer tissues[23].

Interaction with p21WAF1/CIP1

p21 or cyclin dependent kinase inhibitor 1 is a protein which is encoded by the CDKN1A gene located on chromosome 6 (p21.2). The p21 protein binds and inhibits the activity of cyclin-CDK2,
CDK1, CDK4/6; p21 can also mediate cellular senescence. The expression of p21 is controlled by tumor suppressor gene p53\[^{[23]}\]. Several reports have described that p21 concentration can be affected by HPV E5. Tsao \textit{et al} 1996 reported that p21 is repressed by both HPV 11 and 16. They also studied the relation between the transformation activity and p21 suppression effect of HPV by using a series of E5 mutant. In the beginning of their experiment, they used pC11E5 and pC16E5 plasmids which contain an ORF frame of HPV 11 and HPV 16 E5. In the 5' end of this ORFs were cloned with T7 epitope which is a major capsid protein contains a sequence of 11 amino acids. Both plasmids were used the vector pCEP4 and plasmids pC11E5 and pC16E5 and the pCEP4 vector were separately transfected into keratinocytes by Lipofectin transfection with selection for resistance to hygromycin. After 3 weeks, at least 80 hygromycin-resistant colonies were pooled and stable cell lines were generated. They found that cells expressing epitope-tagged HPV-11 E5 and HPV-16 E5 fusion proteins were named Z11E5 and Z16E5. Transforming activity was characterized by anchorage-independent growth. In the beginning of their experiment, they used pC11E5 and pC16E5 plasmids which contain an ORF frame of HPV 11 and HPV 16 E5. In the 5' end of this ORFs were cloned with T7 epitope which is a major capsid protein contains a sequence of 11 amino acids. Both plasmids were used the vector pCEP4 and plasmids pC11E5 and pC16E5 and the pCEP4 vector were separately transfected into keratinocytes by Lipofectin transfection with selection for resistance to hygromycin. After 3 weeks, at least 80 hygromycin-resistant colonies were pooled and stable cell lines were generated. They found that cells expressing epitope-tagged HPV-11 E5 and HPV-16 E5 fusion proteins were named Z11E5 and Z16E5. Transforming activity was characterized by anchorage-independent growth. They revealed that the concentration of p21 RNA and protein were decreased in Z11E5 and Z16E5 cells, which could express HPV-11 and -16 T7-tagged E5 fusion proteins. The decreased concentrations of p21 RNA and protein in cells containing the E5 gene could be due to repression of p21 promoter activity. Next they investigated the promoter activity of p21 gene by transient-transfection assays. Luciferase reporter analysis showed that both HPV-11 E5 and HPV-16 E5 repressed p21 promoter activity in NIH 3T3 cells and keratinocytes\[^{[24]}\].

### Interaction with HLA Class I

HPV infect mucosal and cutaneous epithelia and induce a mildest form of HPV disease, known as warts or papillomas. Most of the cases this warts are removed after several months by the activation of host immune system against HPV's antigen (viral antigen)\[^{[25]}\]. Sometimes certain types of HPVs (mainly HPV 16) avoids the immune clearance and the lesions can progress to cancer (CaCx by HPV 16)\[^{[26]}\]. There are several factors present in HPV life cycle which helps to avoid this immune clearance, for the removal virally infected cells requires cytotoxic T lymphocytes (CTL) which can recognize and kill the infected cells by MHC I (HLA I in humans). HPV 16 E5 protein prevents the transport of major histocompatibility complex or MHC class I to the cell surface and keeps the complex in the Golgi apparatus\[^{[27]}\]. The main effector of HPV 16 is viral oncoprotein E5 which helps to escape from immune clearance. Several report observed that HPV 16 E5 downregulates the expression of surface HLA I by retaining the complex in Golgi apparatus\[^{[28]}\]. In 2006, Ashrafi \textit{et al} worked on the interaction of E5 with the heavy chain of HLA I. At the beginning of their study they constructed HPV 16 E5 mutants by using the plasmid pcDNA-Neo (Invitrogen), encoding G418 resistance and containing the universal IE promoter of CMV. All the wild type and mutant E5 were tagged with hemagglutinin (HA) epitope at their N-terminus. They created four HPV 16 E5 deletion mutants: R79, A54, V36 and R30 by introducing double stop codons at the specific position of nucleotides of HPV 16 E5 sequence. They made HPV 16 E5 Del 1 mutant protein by deletion of first hydrophobic domain by PCR amplification using forward

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**Figure 2** Potential model of HPV 16 E5 ER stress repression\[^{[21]}\].

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primer from nt 91 to 105 of the HPV-16 E5 ORF and reverse primer from nt 252 to 231 including sites for Eco RI and Nhe I. They used pEGFP-C1 eukaryotic expression plasmid which encodes the GFP. They used it for full length cloning of HPV 16 E5 and its mutants to produce GFP-E5 wt and GFP-E5 mutant fusion proteins by PCR. Then they transfected the HaCaT cell line and the RNA was isolated from it and removed the residual DNA by Dnase I treatment, they performed the real-time RT-PCR using Taqman EZ RT-PCR kit for E5, E5-mutant and β-actin and then done a semi-quantitative RT-PCR because semi-quantitative Real time RT-PCR did not work for E5 Del 1 due to lack of appropriate forward primer in the first hydrophobic domain. Next western blotting was performed by Ashrafi et al., they took fifty microgram of protein lysates from HaCaT cell harboring empty vector (Control), expressing Del1 or E5, were electrophoresed and transferred to nitrocellulose membrane. The membranes were incubated with mAb HA 11 for 1 h. and incubated with anti-mouse IgM-HRP and after washing finally the bound antibody was detected by chemoluminescence staining. To detect HLA I, immunofluorescence was performed. At first the cells were washed and fixed in fixing solution which contains PBS, formaldehyde and sucrose, then the cells were incubated in permeabilising solution made by PBS, NP40 and sucrose. Next the cells were incubated with previously mentioned antibodies at 4℃ for 1 h in dark. After 3 final washes with PBS the slides were mounted with glycerol/PBS solution and analyzed under fluorescence confocal microscope. The results of their total experiment were: (1) HPV-16 E5 and HLA I HC interact physically. E5 prevents the transport of the cell surface by retaining the complex in Golgi apparatus[27]. The result of western blotting concluded that E5 and HLA I exist in a physical complex in E5-expressing HaCaT cells; (2) E5 interacts with the HC of different HLA I haplotypes to prove this they performed in-vitro coprecipitation experiments. After this experiments they concluded that the interaction between HC and E5 is likely to take part with most of the alleles of HLA I such as A1, A2, A3, B8 etc. The four deletion mutations shows different characters of E5 mutants, here mutant R79 lacks the last 5C-terminal amino acids, A54 lacks the complete third hydrophobic domain, V36 and R30 lack the 2nd and 3rd hydrophobic domains and Del 1 lacks the 1st hydrophobic domain (Figure 3); (3)The first hydrophobic domain of E5 is responsible for surface HLA I downregulation. They proved it by using flow cytometry, where the level of total (surface+ intracellular) HLA I in HaCaT cells, stable expressed the wild type E5 and its mutants. The clones expressing the E5 mutants containing the first hydrophobic domain (R79, A54, V36, and R30) had reduced level of surface HLA I to the same extent as E5 wt. In contrast, expression of E5 Del1, lacking the first hydrophobic domain, did not have any effect on the levels of surface HLA I. Finally these results conclusively showed that E5 both physically interacts with HC and downregulates surface HLA I via its first hydrophobic domain. In 2007 Gruener et al reported that Calnexin, a molecular chaperone involved in HLA-I trans- port to the cell surface, also physically interacts with this domain of HPV 16 E5 and is required for intracellular retention of HLA-I[41]. In 2010 Campo et al reported that MHC class I is arrested in the Ga by the alkalisation of the endomembrane compartments which is induced by HPV 16 E5, and the direct interaction of E5 with the heavy chain of the MHC class I complex. These conclusions have been reached in cultured HaCaT cells and W12 cell line. The functional effect of the decreased expression of HLA is a reduction of the recognition by CD8+ T cells in vitro[42].

**Interaction with Bax protein**

The role of E6 and E7 has been extensively studied but the role of the E5 protein in cervical carcinogenesis and apoptosis is not clearly recognized[29]. E5 acts as an oncogene primarily and increased the tumorigenicity by enhancing the activation of the epidermal growth factor receptor in a ligand-dependent manner[30]. Apoptosis is a physiological cellular response and one type of programmed cell death. It plays an important role in homeostasis and development[31]. Many viruses, including HPV, have developed various strategies to regulate and block apoptosis and used it as a hallmark of cancer cells[32]. It was already reported that HPV E5 regulate Fas and TRAIL mediated apoptosis and also reported to protect human keratinocytes from UV radiation induced apoptosis by enhancing the phosphatidylinositol 3-kinase-Akt pathway and other signaling pathways[33].

In 2010 Oh et al started to investigate the effect of HPV 16 E5 on apoptosis of cervical cancer cells. They found that the E5 protein inhibits hydrogen peroxide-induced apoptosis by stimulating ubiquitin-mediated degradation of Bax through a cyclooxygenase-2 (COX2) (Figure 4)[40], prostaglandin E2 (PGE2), EP2, EP4 and cyclic adenosine monophosphate-dependent protein kinase-dependent pathway in C-33A human cervical cancer cells. They analyzed the effect of HPV 16 E5 on H2O2 induced apoptosis using the C-33A human cervical cancer cells[41]. They confirmed the expression of E5 mRNA and protein by RT-PCR and western blot analysis. They performed a double immunofluorescence analysis using a confocal microscope to examine the cellular location of FLAG-E5 protein and found the E5 protein was colocalized with a 58K cellular protein. The effect of E5 on hydrogen peroxide-induced apoptosis was analyzed by caspase-3 cleavage and cytochrome c release using western blot analysis. E5 expression decreased the cleavage of caspase-3 from 10.1+1.2 fold and the cytochrome c released using western blot analysis. They also found that HPV16 E5 decreases the expression of pro-apoptotic Bax and Bak but increases the expression of anti-apoptotic Bcl-2 by western blotting. They exposed the Bax protein for a long time in the blot to film and the expression of Bax protein was decreased significantly where the level of Bcl protein was not changed by E5 expression. They further examined the role of Bax in the anti-apoptotic action of E5, the effect of Bax expression on the hydrogen peroxide-induced apoptosis of C-33A cells and they observed the level of Bax expression was decreased where anti-apoptotic Bcl-2 level was increased slightly in C-33a cells. These results suggest that E5 may exert an anti-apoptotic effect by changing the expression of Bax, Bak and Bcl-2 proteins in cervical cancer cells and E5 inhibits hydrogen peroxide-induced apoptosis partly by decreasing Bax expression in cervical cancer cells. They simultaneously studied the Bax mRNA level by RT-PCR.
and efficiency of Bax promoter by Bax-luciferase activity but the Bax mRNA level was not decreased significantly in E5-expressing C-33A cells and also the Bax promoter activity was not decreased. This result indicates that E5 does not inhibit the transcription of Bax but the inhibition and regulation starts during post-translational modification and change the Bax expression at protein level. Next they examined the effect of PGE2-signaling pathway on Bax-degradation by E5 protein. Treatment with PGE2 decreased hydrogen peroxide-induced apoptosis of E5-transfected C-33A cells and also decreased the Bax expression. This result suggests that E5 inhibits hydrogen peroxide induced apoptosis by stimulating PGE2-signaling pathways to downregulate Bax expression in C-33A cells. From this study they concluded that HPV E5 protein inhibits hydrogen peroxide-induced apoptosis mediated by COX-2, PGE2: which degraded the Bax protein in cervical cancer cells.

Some recent studies on this topic said that HPV 16 E5 on COX-2 expression demonstrated that E5 upregulates COX-2 expression through the EGFR signaling pathway, with nuclear factor-kappa B (NF-kB) and AP-1 as critical factors.

**FUTURE DIRECTIONS**

In this review article I have tried to discuss the role of HPV 16 E5 protein in CaCx and some future perspective of E5 which will make this protein a major oncogene of HPV and potential biomarker of HPV infected CaCx. Oncogenic high-risk HPV types induce malignant transformation in cervical mucosal epithelia by expressing E5, E6 and E7 oncoproteins and maintains it malignant phenotype by continuous expression of E6 and E7 proteins. But the overall role of E5 oncoprotein in cancer is less understood still now. So, there are some areas to work on HPV E5 protein in future and they are: (1) To analyze the random effects of E5 protein in triggering malignant changes in epithelial cells; (2) To examine the cellular levels alterations in gene and protein expression due to expression of HPV E5 in epithelial cells, identify the altered gene by microarray platforms; (3) To evaluate the effects of E5 on cellular microRNAs & study their changes in gene expression; (4) To reveal the role of E5 on cell adhesion and cell motility, epithelial to mesenchymal transitions (EMT) cellular signal transduction and transformations and finally carcinogenesis.

**CONFLICT OF INTERESTS**

There are no conflicts of interest with regard to the present study.

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Peer reviewers: Kiyoshi Yoshino, M.D., Ph.D, Department of Obstetrics and Gynecology, Osaka University, Graduate School of Medicine, 2-2, Yamadaoka, Suita, Osaka 5650871 Japan; Yutaka Ueda, Assistant Professor, Department of Obstetrics and Gynecology, Osaka University Graduate School of Medicine, 2-2 Yamadaoka, Suita, Osaka, 565-0871, Japan; Eliane Campos Coimbra, Universidade Federal de Pernambuco - UFPE, Departamento de Genética, Laboratório de Estudos Moleculares e Terapia Experimental-LEMTE, Av. Prof. Moraes Rego, 1235, Cidade Universitária, Recife- Pernambuco, Brasil.