Contents

List of abbreviation:

DNA	Deoxyribonucleic Acid
HPV	Human Papillomavirus
URR	Upstream Regulatory Region
LCR	Long Control Region
E	Early region
L	Late region
PRb	Retinoblastoma Protein
BPV-1	Bovine Papillomavirus type 1
VLPs	Virus-like Particles
E6AP	E6-Associated Protein
Cdk	Cyclin dependent kinase
CDP	CCAATT Displacement Protein
ORF	Open Reading Frame
RNA	Ribonucleic Acid
mRNA	Messenger Ribonucleic Acid
CPSF	Cleavage and Plyadenylation Specificity Factor
SD	Splice Donor
SA	Splice Acceptor
BM	Basement Membrane
HSPGs	Heparin sulfate Proteoglycans
TGN	Trans-Golgi-Network
ER	Endoplasmic Reticulum
ECM	Extracellular Matrix
KIK	Kallikrein
PC	Proprotein Convertases
MMP	Matrix Metalloproteases
GF	Growth Factor
GFR	Growth Factor Receptor
AZt	Annexin AZ tetramer
APC	Antigen presenting cells
DC	Dendritic cells
CTL	Cytotoxic T cell
Tneg	Regulatory T cell
Pap	Papanico_laou
STI	Sexually Transmitted Infection
LEEP	Loop Electrosurgical Excision Procedure
VIA	Visual Inspection with Acetic acid
VILI	Visual Inspection using Lugol's Iodine
WHO	World Health Organization

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1. Introduction :

Human Papillomavirus (HPVs) are a family of deoxyribonucleic acid (DNA) viruses that infect skin or mucosal epithelial cells. Of the more than 100 types of HPV, at least 13 can cause cancer of the cervix, other anogenital organs, or the head and neck. Other HPV types with low oncogenic potential cause non-malignant lesions, such as anogenital warts [1]. HPV types 16 and 18 are responsible for about 70% of all cervical cancer cases worldwide[2]. Cervical cancer is the most common HPV-related malignancy. It is the leading cause of cancer among women in developing countries, and the second leading cancer in women worldwide. In 2005, nearly 500 000 new cases of cervical cancer occurred. About 80% of cervical cancer deaths occur in developing countries; if current mortality trends continue, this proportion is expected to increase to 90% by 2020 [1]. Although it is difficult to estimate the overall prevalence of genital human papillomavirus (HPV) infection, current figures suggest that visible genital warts are present in approximately 1% of sexually active adults in the United States and that at least 15% have subclinical infection, as detected by HPV DNA assays. Genital HPV infection is thus extremely common. The highest rates of genital HPV infection are found in adults 18–28 years of age. Although risk factors for infection are difficult to assess because of the high frequency of subclinical infection, it is clear that major risk factors for acquiring genital HPV infection involve sexual behavior, particularly multiple sex partners [3]. However, a number of infected women do not develop invasive lesions, suggesting that other environmental and host factors may play decisive roles in the persistence of HPV infection and further malignant conversion of cervical epithelium [4].

2. About the virus :

2.1Introduction of the virus

Human papillomaviruses (HPVs) are small double-stranded DNA viruses that induce hyperproliferative lesions of cutanous and mucosal epithelia. Of the more than 100 identified types of HPVs, approximately 30 specifically infect the genital epithelium [5]. Genital papillomavirus infections can be passed between individuals through sexual contact, and represent one of the most common sexually transmitted diseases. Some HPV types like 6 and 11 induce only benign warts in the genital tract (condylomata acuminatum). They are referred

to as low-risk types, as they induce lesions at low-risk for progression to malignancy. In contrast, the high-risk HPV types are associated at a high frequency with the development of malignant lesions. Approximately 99% of cervical cancers contain HPV DNA of the high-risk types [6].

2.2Classification of the virus

Papillomaviruses are classified as genus Papillomavirus of the Papovaviridae family on the basis of their capsid structure and biochemical composition. Polyomalike viruses form the second genus and are distinguished by a smaller capsid and a shorter DNA, Both genera reveal group-specific antigens and the nucleic acids of individual members hybridise under conditions of reduced stringency. However, there ist no cross-reactivity between papillomaviruses and polyomaviruses. DNA sequen analysis has disclosed a fundamentally different genome organization. There is only one coding DNA strand in papillomaviruses whereas separate strands code for vegetative functions and structural proteins in polyomaviruses. This indicates that papillomaviruses represent a distinct group [7].

2.3Structure and genome

The papilloma viruses are small 52-55 nm in diameter and non-enveloped double strand of DNA viruses encased in 72sided icosahedral protein capsid. The HPV genomes consist of circ ular double stranded DNA approximatly of 7.900 nucleotide base pair this shape is made up of 12 pentamerric and 60 hexameric arranged on a T=7 lattice their capsid is composed of two proteins a major (L1) and minor(L2) [8].

2.4Protein (Virulence Factors)

The HPV genome is divided into three regions. The first is a noncoding upstream regulatory region (URR) or long control region (LCR) that has regulatory function of the transcription of the E6 and E7 viral genes; The second is an early region (E), consisting of six ORFs: E1, E2, E4, E5, E6, and E7, which encodes no structural proteins. The third is a late (L) region that encodes the L1 and L2 structural proteins [9].

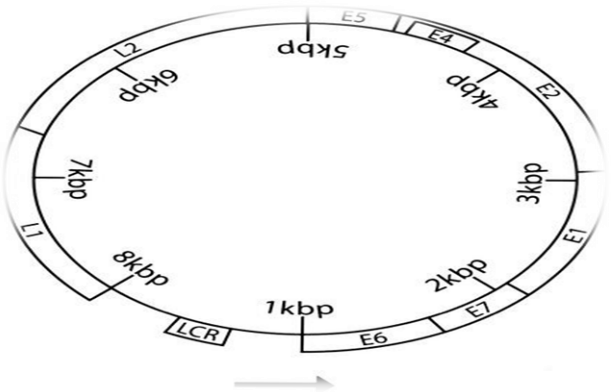


Figure1: Schematic representation of the circular HPV DNA genome.

2.4.1 Structural proteins and their function:

There is two type of structural proteins : L1(major protein) and L2(minor protein). L1 Major capsid protein; contains the major determinant required for attachment to cell surface receptors. It is highly immunogenic and has conformational epitopes that induce the production of neutralizing type-specific antibodies against the virus[10]. L2 Minor capsid protein; L2 contributes to the binding of virion in the cell receptor, favoring its uptake, transport to the nucleus, and delivery of viral DNA to replication centers. Besides, E2 helps the packaging of viral DNA into capsid[11] .

2.4.2 non- Structural proteins:

There is many types of non-structural proteins : E1, E2, E4, E5, E6, and E7. E6 and E7 proteins of especially high risk HPV types (e.g. types 16 and 18) interact with tumor supressor proteins such as p53 and retinoblastoma (pRb) proteins, respectively; inhibit their functions and cause uncontrolled proliferation and immortalization of the cells. The binding of E6 protein to p53 leads its rapid degradation, and DNA repair mechanisms and apoptosis are terminated. In the other way, E7 protein interacts with pRb and mitotically interactive cellular proteins such as cyclin-E, causing stimulation of cellular DNA synthesis and cell proliferation finally leading to cervical cancer [12].

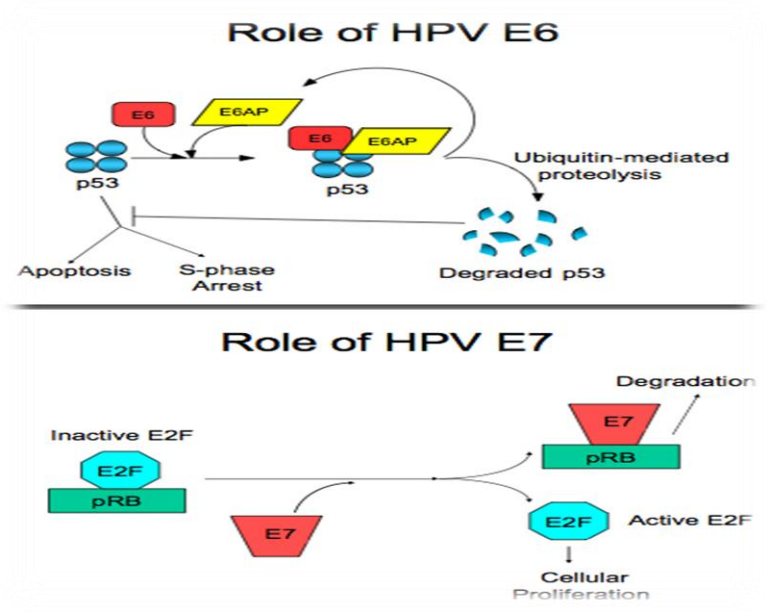


Figure2: The role of E6 and E7 proteins to degradation of tumor suppressor protein p53 and Rb leading to uncontrolled activation of cell proliferation.

3. Replication cycle :

Attachment, Entry, and Uncoating

The first step in viral infection is the binding of the virion to the cell. Binding studies with radiolabeled virions have revealed that the papillomaviruses can bind a wide variety of cell types in addition to the normal host cell, the squamous epithelial cell. The specific tropism of these viruses for keratinocytes does not appear to be due to a cell-type–specific receptor. This observation is consistent with studies with the BPV-1, which have shown that this virus can infect and transform a wide variety of cells, including rodent fibroblasts. The receptors by which papillomaviruses bind and enter cells have not been unequivocally identified. The α6 integrin was first identified as a candidate receptor for the papillomaviruses based on studies showing that papillomavirus VLPs bound a protein that was identified as a6 and that antibodies to α6 could block binding of VLPs to cells . Recent studies have established that the expression of the α6 integrin in a receptor-negative cell line is sufficient to confer papillomavirus binding to the cell, providing functional evidence for the role of a6 as the receptor The α6β1 integrin is expressed on a wide range of cells, including platelets, lymphocytes, endothelial cells, and so forth, whereas α6β4 is on relatively restricted epithelial cells, mesenchymal cells, and some neuronal cells. Papillomaviruses can bind cells expressing either the α6β1 or the α6β4 complex, although the cells with the highest degree of papillomavirus binding match the α6β4 expression profile. The virions can also bind to

heparin and cell surface glycosaminoglycans on human keratinocyte.The virus particles were transported in phagosomes, and their uptake and transport could be inhibited by cytochalasin B and paclitaxel (Taxol), suggesting the possible involvement of microfilaments and microtubules in these processes. Although binding to the plasma membrane and uptake of virions into large cytoplasmic vesicles could be monitored by electron microscopy, no complete virions were observed in the nucleus of infected cells This observation suggests that disintegration of the virion occurs in the cytoplasm and that the L1 and L2 proteins may migrate to the nucleus through their nuclear localization signals. despite a very strong nuclear fluorescent staining for both L1 and L2 proteins

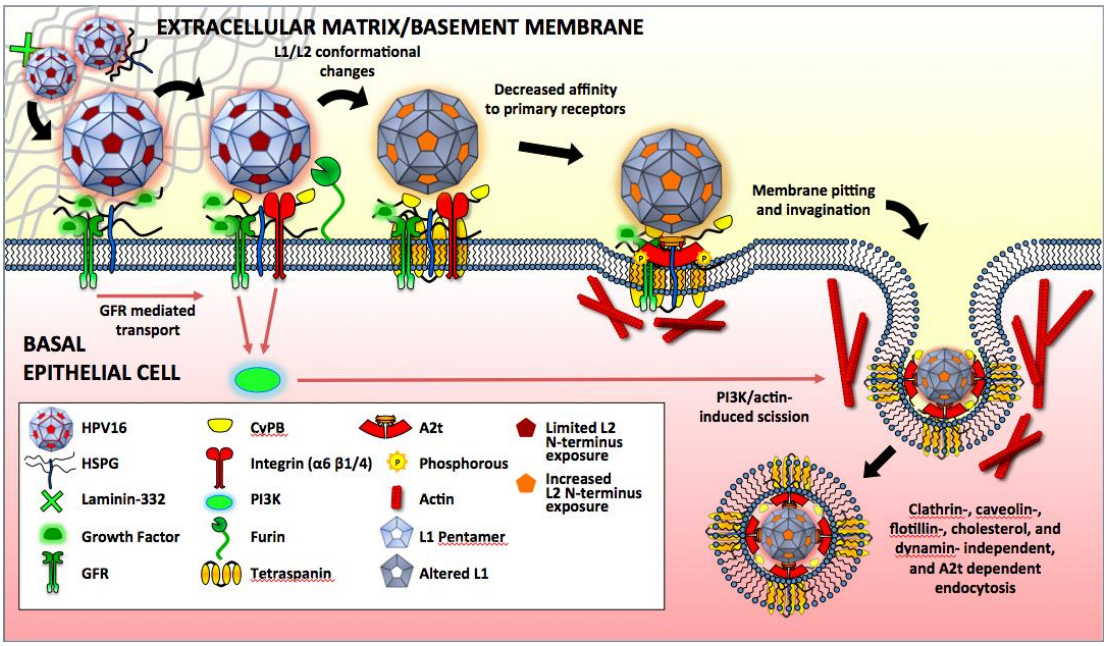


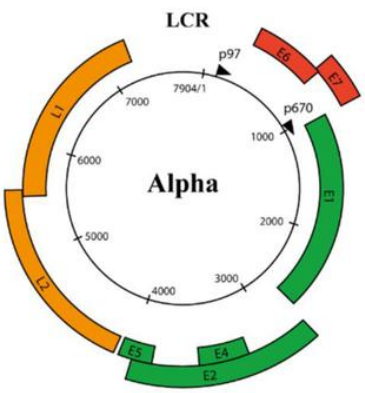
Figure3. Attachment, Entry, and Uncoating of HPV to cells

3.1HPV Replication:

The papillomaviruses are highly species specific, induce squamous epithelial tumors and fibroepithelial tumors in their natural hosts, and have a specific tropism for squamous epithelial cells. The productive infection of cells by the papillomaviruses can be divided into early and late stages. These stages are linked to the differentiation state of the epithelial cell [13]. The HPV genome contains eight genes with multiple promoters and a number of splice variants that are expressed either early (E) or late (L) in the HPV lifecycle. The early genes encode nonstructural proteins that participate in DNA replication,transcriptional regulation, cell transforma tion and viral assembly and release. The late (L) genes, L1 and L2, encode viral capsid proteins. An additional 1000bp noncoding region of the 8000bp HPV genome contains transcriptional regulatory sequences and the viral origin of replication [14]. The specific tropism of the papillomaviruses for squamous epithelial cells is evidenced by the restriction of the viral replication functions, such as vegetative viral DNA synthesis, the production of viral capsid proteins, and the assembly of virions, to keratinocytes in the

process of terminal differentiation. The close link of the papillomavirus life cycle with the differentiation program of the squamous epithelium is depicted in [15]. The first viral genes to be expressed, E6 and E7, are involved in cell transformation in HPV high-risk types. E6 complexes with the cellular ubiquitin ligase. The E6AP and the E6AP/E6 complex acts as a p53 specific ubiquitin protein ligase to increase the rate of p53 degradation, thereby negating the anti-proliferative and pro-apoptotic functions of p53 in cases of DNA damage and cellular stress[16] . The early gene regions of the papillomaviruses, viral transcripts have been detected in the basal cells of the epidermis . some early viral protein is found in basal cells[17]. E7, similar to E6, is also an oncoprotein. E7 complexes with hypophosphorylated pocket proteins pRb, p107 or p130, facilitating the release of the transcription factor E2F, which then constitutively activates DNA synthesis and cell proliferation [18]. The Rb tumor suppressor protein normally binds to and inactivatesE2F. E7 has also been shown to inactivate .the cyclin dependent kinase (Cdk) inhibitors,p21/Cip1 and p27/Kip1 and complex with cyclins A and E Late gene expression, synthesis of capsid proteins [19].The late promoter is specifically activated in the differentiated layers of epithelium. The late promoter activity is suppressed by CDP (CCAAT displacement protein) and YY-1, whose binding potential was reported to be decreased in differentiated keratinocytes [20]. the late region (L), encodes L1 and L2 structural proteins that form the major and minor capsid proteins, respectively [21] . There was also a report that the expression ratio of a transcription factor, Sp1 and its antagonist, Sp3, was altered through the differentiation, which activated the late promoter activity [22].

A



B

Core Proteins	
E1	ATP- dependent helicase. Role in papillomavirus genome replication.
E2	Coactivator of viral genome replication through the recruitment of E1 to the viral replication origin. Transcription factor of E6 and E7, also important for viral genome segregation.
L1	Major capsid protein. Assembles into pentomeric capsomeres, which are the primary components of the icosahedral virion shell.
L2	Minor capsid protein. Involved in encapsidation of viral DNA. Facilitate virus entry and trafficikng to the nucleus.
E4	E4 gene is embedded within the E2 gene, and is expressed abundantly as an E1^E4 fusion protein during the late stages of the viral life cycle. Binds to cytokeratin filaments and disrupts their structure. E4 is thought also to contribute to virus release and transmission.
Accessory Proteins	
E5	Small transmembrane protein. In Alpha-PVs, E5 interacts with the EGF receptor and activates mitogenic signaling pathways. Has a role in evading the immune response and apoptosis. Beta-, Gamma- and Mu-PVs lack E5 gene.
E6	Drives cell cycle entry to allow genome amplification in the upper epithelial layers. E6 of high-risk Alpha-PV types binds and degrades p53 and can also activate telomerase and contribute to transformation. Also involved in immune evasion. Some Gamma-PVs lack E6 gene.
E7	Drives cell cycle entry to allow genome amplification in the upper epithelial layers. E7 of high-risk Alpha-PV types binds and degrades pRb and can induces chromosomal instability. E7 is necessary for cell transformation. Both E6 and E7 have a large number of cellular substrates, with the identity of these substrates differing between HPV types [78].

Figure 4. Alphapapillomavirus genome organization and the function of HPV proteins. (A) Genome organization typical of the high-risk Alphapapillomavirus types is illustrated by the genome of HPV16. The early (p97) and late (p670) promoters are marked by arrows. The six early ORFs (E1, E2, E4 and E5 (in green) and E6 and E7 (in red)) are expressed from the different promoters at different stages during epithelial cell differentiation. The late ORFs (L1 and L2 (in yellow)) are expressed from the p670 promoter in the upper epithelial layers as result of changes in splicing. The LCR/URR also contains the replication origin as well as post-transcriptional control sequences that contribute to viral gene expression. (B) The function of viral proteins. All known papillomavirus encodes a group of "core" proteins that were present early on during papillomavirus evolution, and which are conserved in sequence and in function between PV types. These include E1, E2, L2 and L1. The E4 protein may also be a core protein that has evolved to meet papillomaviruses epithelial specialization. The accessory proteins have evolved in each papillomavirus type during adaptation to different epithelial niches. The sequence and function of these genes are divergent between types. In general, these proteins are involved in modifying the cellular environment to facilitate virus life cycle completion, contributing to the virulence and pathogenicity. Knowledge of accessory protein function comes primarily from the study of Alphapapillomavirus types.

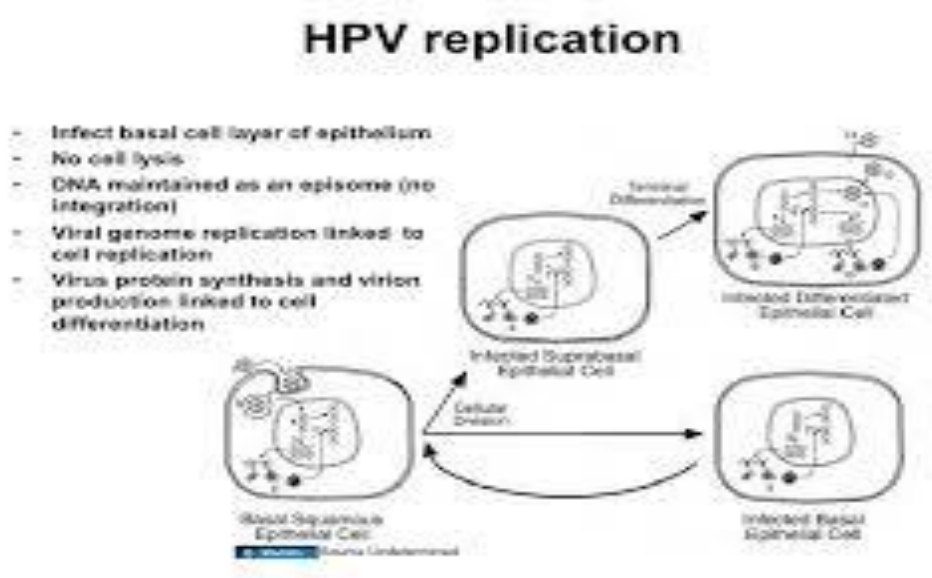


Figure 5. Replication cycle of a papillomavirus. To establish a wart or papilloma, the virus must infect a basal epithelial cell. Our knowledge is limited about the initial steps in the replication cycle such as attachment (1), uptake (2), endocytosis (3), and transport to the nucleus and uncoating of the viral DNA (4). Early-region transcription (5), translation of the early proteins (6), and steady-state viral DNA replication (7) all occur in the basal cell and in the infected suprabasal epithelial cell. Events in the viral life cycle leading to the production of virion particles occur in the differentiated keratinocyte: vegetative viral DNA replication (8), transcription of the late region (9), production of the capsid proteins L1 and L2 (10), assembly of the virion particles (11), nuclear breakdown (12), and release of virus (13).

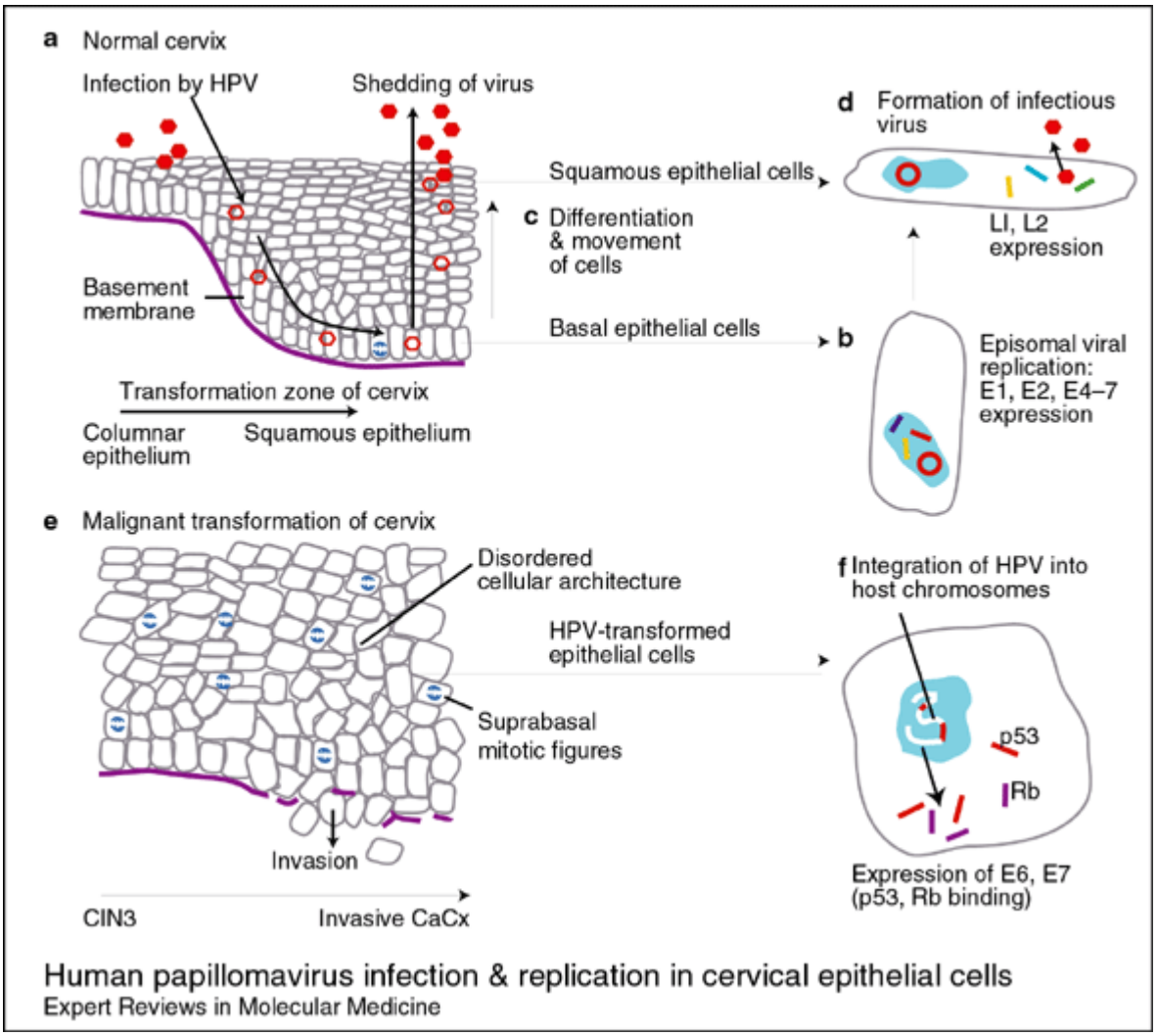


Figure 6.Human papillomavirus (HPV) infection and replication in cervical epithelial cells. (a) The normal cervix has a (narrow) transformation zone in which there is an abrupt transition from a columnar epithelium (sometimes via a metaplastic epithelium) to a squamous epithelium; HPVs are probably most infectious to cells that are close to this junction. (b) HPV viruses gain access to the basal epithelial cells of the cervix via the vagina (for example, during sexual intercourse), where they replicate episomally (outside the host chromosome in the nucleus) and express the (early) viral genes E1, E2, E4, E5, E6 and E7. (c) The infected basal cells, which show signs of cell disruption as a result of the viral infection, continue their differentiation and migration to the epithelial surface, where (d) the (now) squamous cells start to express the late HPV genes LI and L2. Infectious virus particles are formed and shed into the lumen of the vagina. (e) HPV infection (particularly with the high-risk types) can progress to: (1) HPV-induced mild dysplasia, (2) the final stages of cervical intraepithelial neoplasia (CIN3) and, eventually, (3) invasive cervical cancer (CaCx), when the basement membrane is breached by the cells, allowing local spread and also distant metastasis. (f) In transformed epithelial cells, HPV genes are integrated into the host chromosomes, with expression of (the oncogenic) E6 and E7 proteins, which bind to the tumour-suppressor proteins p53 and Rb

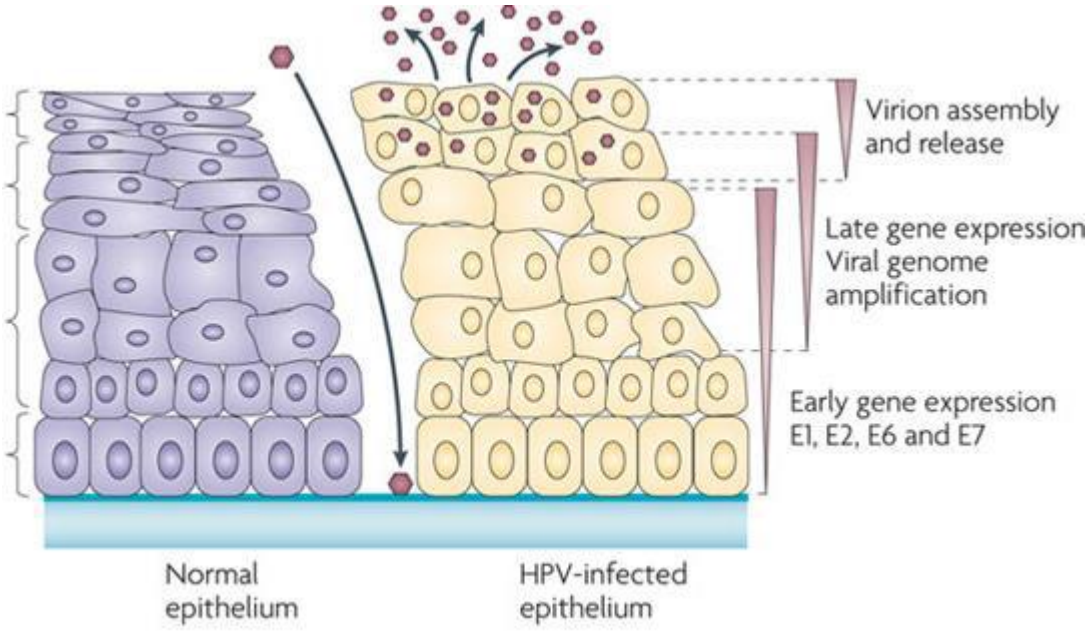


Figure 7. The Life Cycle of Human Papillomaviruses Uninfected epithelium is shown on the left and HPV-infected epithelium is shown on the right. HPV gains entry to the basal cell layer of the cervical epithelium through micro-abrasions. Once initial infection has occurred, the early HPV genes E1, E2, E6 and E7 are expressed and the viral DNA replicates from episomal DNA. As the viral genome migrates towards the upper epithelium, it is further replicated and the late genes, L1 and L2, as well as E4, are expressed. These L1 and L2 proteins allow the viral DNA to become enclosed into capsids and form virions. which are the shed from the cell .

3.2Gen Expression

Gene expression is the method by which genetic instructions are used to synthesise gene products. The process of gene expression involves two key stages i.e. transcription and translation. Transcription involves the production of messenger RNA (mRNA) from DNA by the enzyme RNA polymerase and the processing of the resulting mRNA molecule. Translation refers to the subsequent use of this mRNA to direct protein synthesis, and the successive posttranslationalprocessing of the protein molecule [23]. The first step in transcription in the cytoplasm is the formation of pre-mRNA from a DNA template. This pre-mRNA must undergo three major processing events, referred to as capping, splicing and polyadenylation,before it can become a mature and stable mRNA and be exported to the translation machinery [24].

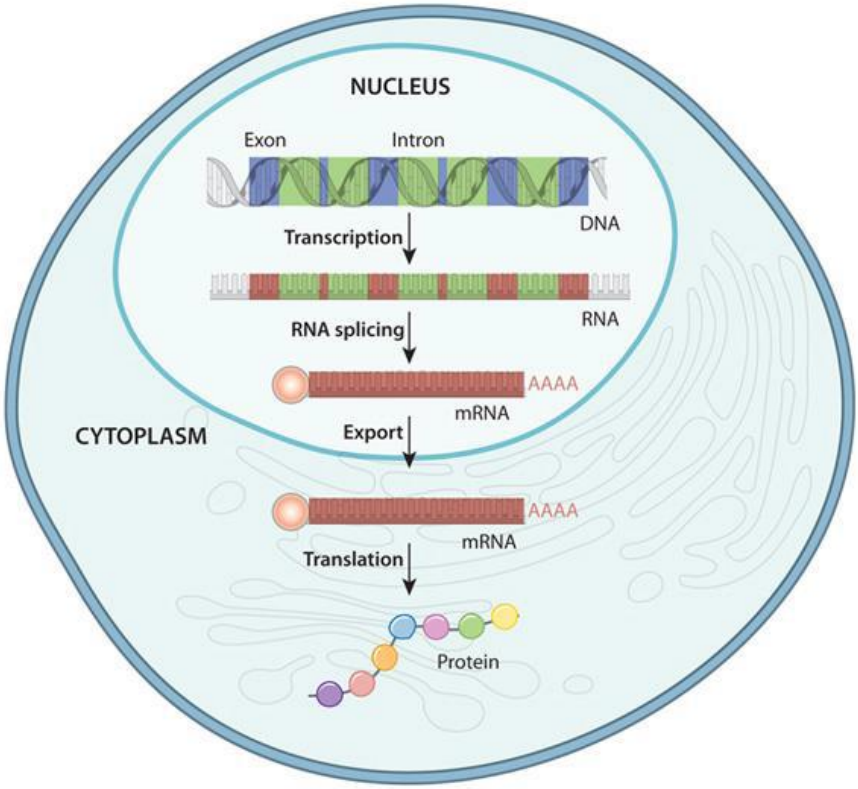


Figure 8. mRNA Processing The processing of pre-mRNA includes capping, splicing and polyadenylation before eventual construction of a protein.

Capping involves the addition of 7-methyl guanosine groups (mRNA "cap") to the 5′ ends of the newly synthesized pre-mRNA. This occurs once approximately 20-30 nucleotides of the molecule have been transcribed and requires removal of the terminal 5' phosphate, which is achieved with the aid of a phosphatase enzyme. The process of capping converts the 5’ end to a 3’ end by 5'-5' linkage, protecting the mRNA from 5' exonuclease, which degrades foreign RNA. The newly formed complex assists with the binding of ribosomes to the mRNA during translation and also aids in the protection of the mRNA from premature degradation [25].

3.3Splicing

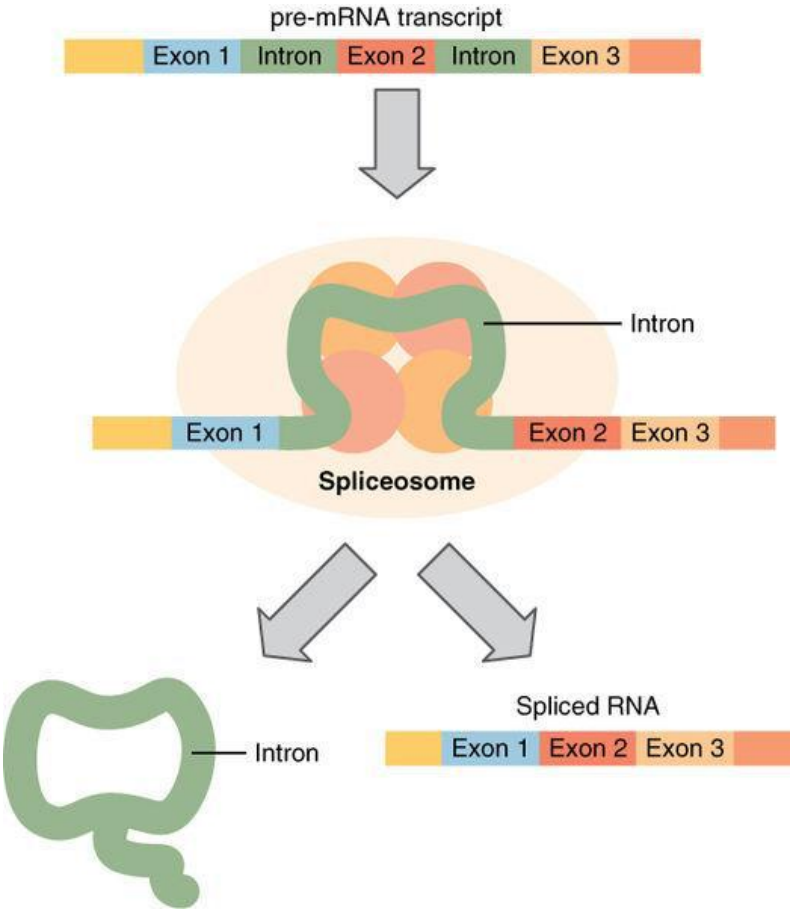


Figure 9.The figure illustrates the exons and introns in pre-mRNA and the formation of mature mRNA through the removal of noncoding introns as occurs with splicing.

Polyadenylation is a method utilised in gene regulation in which a sequence of adenosine ribonucleotides are added to the 3' end of a spliced mRNA to form a poly(A) tail. The primary transcript is cleaved at the polyadenylation signal sequence, an AAUAAA sequence, by the cleavage and polyadenylation specificity factor (CPSF). The poly (A) tail is a useful tool in the protection of mRNA from digestion with nuclease and greatly increases the efficiency of translation [26].

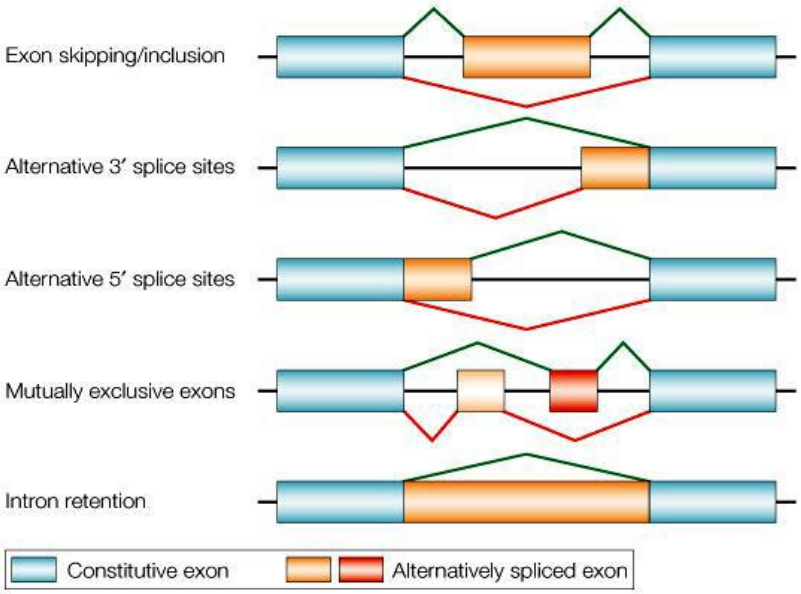


Figure 10.Alternative Splicing The diagram shows different types of alternative splicing including exon inclusion or skipping, alternative splice-site selection, mutually exclusive exons and intron retention.

Expression of the human papillomavirus capsid genes, L1 and L2, as well as amplification of viral DNA and virion assembly occur only in the terminally differentiated layers of infected epithelium. Furthermore, it has also been established that HPV-16 late genes are not expressed in cervical cancer containing HPV-16 DNAHPV Late Gene Expression. Eleven splice sites have also been identified, 10 located in the early region and only 1 in the late region, as displayed in [27] E4 mRNA is one of the most abundant HPV-16 mRNAs produced and is generated from splice donor (SD880) to splice acceptor (SA3358). The most efficient splice site utilised by HPV-16 is this major 3'-splice site SA3358, which is involved in the production of E4, E6, E7, L1 and L2. Late mRNAs are transcribed from the late promoter and are thought to be spliced either from SD880 to SA3358 and from SD3632 to SA5639 or directly from SD880 to SA5639. The SD3632 and SA5639 are used exclusively by the late mRNAs and the presence of an adjacent splicing silencer that actively suppresses the use of these splice sites has been shown to inhibit late gene expression [28] a productive HPV-16 infection and is also likely to be important for pathogenesis. Alternative splicing appears crucial for the production of L1, since the 3′ end of L2 and the 5′ end of L1 overlap. As mentioned, late viral mRNAs are expressed only in differentiating cells and it has been demonstrated that posttranscriptional events are highly involved in late gene regulation [21] These proteins are a source of specific interest, as, should a method be determined to induce their expression, late gene products could be subsequently up-regulated [29].

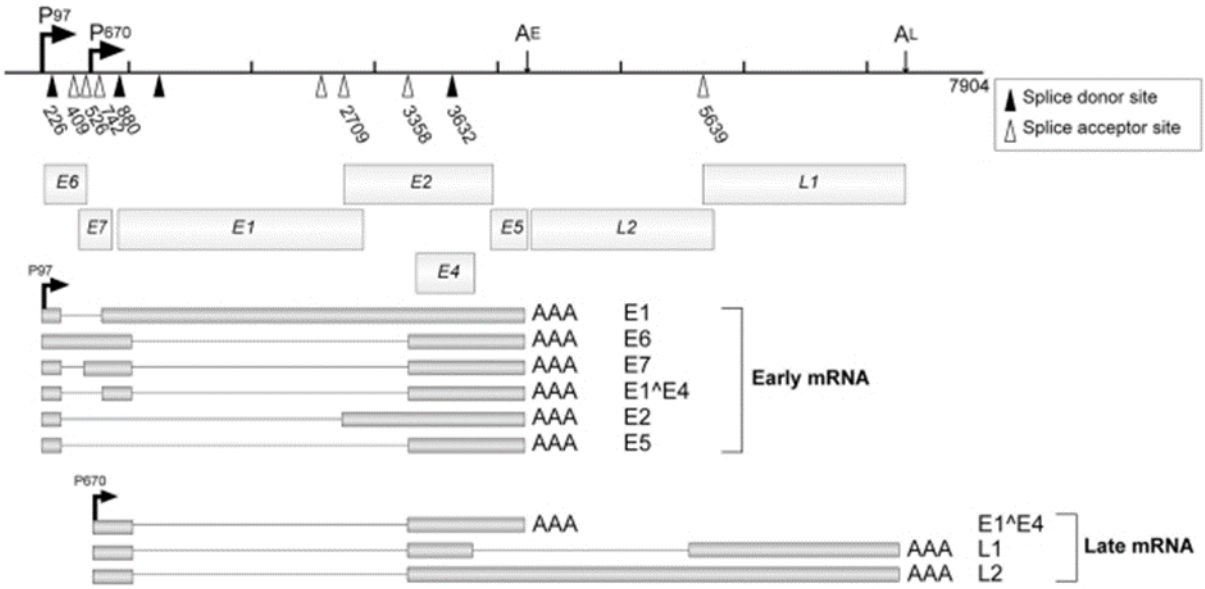


Figure 11. The splicing signals and the transcripts of HPV16. P97 and P670 are the early and late promoters, respectively, for HPV16. AE and AL indicate the early and late polyadenylation signals, respectively. Open triangles indicate splicing acceptors and filled.

4. Transmission

You can have the genital HPV virus for years and not have any sign of it. There is no way of knowing how long you have had the virus. It could be weeks, months or years. Genital HPV is usually spread through intimate, skin to skin, contact during sex. The risk increases with the number of sexual partners a person has To give examples, all HPV types that form a species together with HPV-16 are ‘‘high-risk’’ HPV types found in cervical cancer and its precursor lesions, and all HPV types that form a species together with HPV-2 are typically found in common skin warts, Those anogenital warts are caused by HPV [1].

4.1Types of HPV transmission

4.1.1 Horizontal transmission:

The most common mode of horizontal transmission of anogenital HPV is by sexual activity through contact with infected cervical, vaginal, vulvar, penile or anal epithelium.

4.1.2 Vertical transmission:

IS infection that transmitted by nonsexual routes, but this appears to be uncommon. Nonsexual routes of genital HPV transmission include transmission from a woman to a newborn infant at the time of birth.

4.1.3 Issues in assessing transmission:

The types of HPV that affect the skin can be passed on by skin contact with an affected person. The types of HPV that affect the mouth and throat can be passed on through certain sexual behaviours such as open mouth kissing and oral sex , also couples with genital-to-genital transmission reported vaginal intercourse during the period corresponding to the transmission event. Among 5 from 25 couples with penisto- anus transmission. Transmitting couples had more frequent sexual intercourse with one another, were more likely to have contact between the male's mouth and the female's anus, were more likely to use birth control injections and have withdrawal before ejaculation, and had fewer periods of abstinence. Over half of nontransmitting couples reported use of condoms 100% of the time during sexual intercourse within the previous 4 months, compared with only 3% of transmitting couples.The mean age 28 years (range 18–59 years) for men and 26 years (range 18–57 years) for women. Participants comprised Caucasians (52%) [1, 30].

5. Penetration and target organ

HPV16 infection is a multistep process: the virus takes advantage of a trauma to the epithelium to reach the basal keratinocytes—the only mitotically active cells of the epithelium. In vivo, the virus first binds to the basement membrane (BM) and then to the cell surface through interactions between the viral L1 protein and heparin sulfate proteoglycans (HSPGs). Cellular entry requires several conformational changes within the capsid via proteases and chaperones, and interaction of the capsid proteins with different receptors. The virus interacts with an elusive receptor complex and is subsequently internalized via a non-traditional endocytosis mechanism. During intracellular trafficking, the virus has been shown to localize to the endosomal system, trans-Golgi-network (TGN), Golgi complex and endoplasmic reticulum (ER) before delivery to the nucleus, where the viral DNA replicates [31]. The virus targets epithelial cells of the cervix [9].

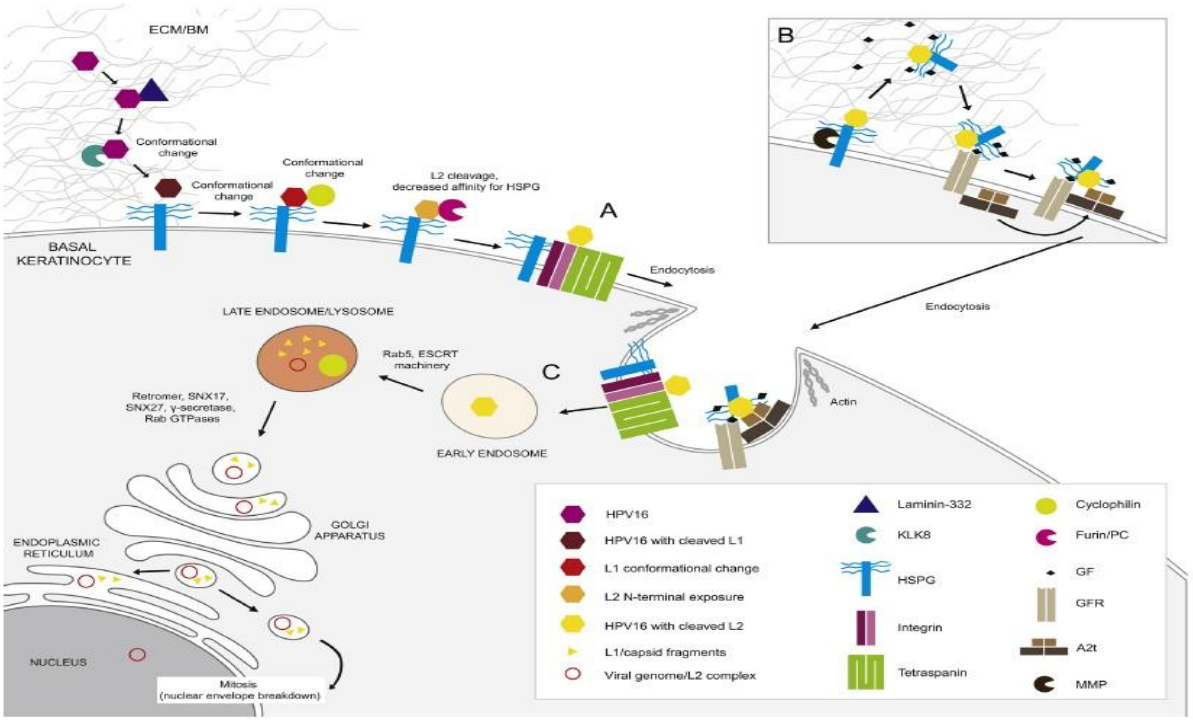


Figure 12. Our current understanding of HPV16 binding, entry, and trafficking during infection of a basal keratinocyte. In the extracellular matrix (ECM) or (BM), HPV16 transiently binds to Laminin-332. Kallikrein-8 (KLK8) cleaves the L1 capsid protein causing a conformational change in the viral capsid. HPV16 binds to (HSPGs) on the cell surface. Conformational changes expose the N-terminal region of L2 in the viral capsid, and may be facilitated by cyclophilins. Furin/ proprotein convertases (PCs) cleave the exposed N-terminus of L2, causing decreased affinity of the virus for the HSPGs. This allows for movement of the virus to an uptake receptor complex, whose identity is unknown. A) Integrins and tetraspanins are candidates for the uptake receptor complex. B) Alternatively, HSPGs with bound virus can be cleaved by matrix metalloproteases (MMPs) during normal cell surface HSPG turnover, which results in virus-HSPG complexes being shed into the ECM. A growth factor (GF)/ HSPG/HPV16 complex can subsequently interact with growth factor receptors (GFRs) on the cell surface. Activation of GFRs can cause the translocation of Annexin A2 tetramer (A2t) from the inner leaflet of the plasma membrane to the outer leaflet. The HPV16/HSPG/GFR/A2t complex can then be internalized by the cell. C) Endocytosis occurs via a macropinocytosis-like mechanism involving actin. The virus traffics through the endolysosomal system where L1 mostly dissociates from the viral genome/L2 complex, facilitated by cyclophilins. The viral genome/L2 complex traffics through the Golgi and may enter the ER, or be in an ER cisternae, before gaining access to the nucleus when the nuclear envelope breaks down during mitosis [31]

6. Genetics (Gene Mutation):

HPV infects actively well-differentiated keratinocytes at the basal epithelial layer of the stratified squamous epithelium. It integrates into the host genome randomly and encodes for six non-structural viral regulatory proteins (E1, E2, E4, E5, E6 and E7) from the early region of the viral genome and two structural viral capsid proteins (L1 and L2) from the late region. The DNA integration process is preceded by the disruption of the E1/E2 regions, with the further deletion of the E2 region. As a consequence, E6 and E7 promoters are activated; E6 is the earliest expressed gene during HPV infection [32, 33]. Only the early genes are transcribed after viral DNA replication. E6 binds with the tumor suppressor gene p53, inducing its degradation, which results in prevention of apoptosis [34, 35]. E7 has numerous interactions with cellular proteins involved in cell growth regulation, such as cyclin-dependent kinases (CDKs) and CDK inhibitors, but it interacts particularly with retinoblastoma suppressor protein (Rb) by binding to the G0/G1-specific hypophosphorylated form of Rb, disrupting the pRb/E2F complex and bypassing cell cycle arrest [32, 36, 37]. E5 protein binds with the platelet-derived growth factor β receptor, promoting a sustained mitogenic signal [38]. These events are synergistic and lead to deregulation of cell growth with the convenient inhibition of apoptosis, which allows for the accumulation of mutations [39].

7. Host Immune Defense

First, HPV can infect cells through damaged skin tissue. When the damage reaches the basal layer of the epidermis, the virions can infect dividing keratinocytes.
The initial inflammatory response induced by tissue damage leads to infiltration of immune cells mainly neutrophils, followed by macrophages and later lymphocytes. These nonspecific innate immune cells detect “danger” by recognizing viral molecules, such as the double-stranded DNA HPV genome or the L1 and L2 capsid proteins [40].

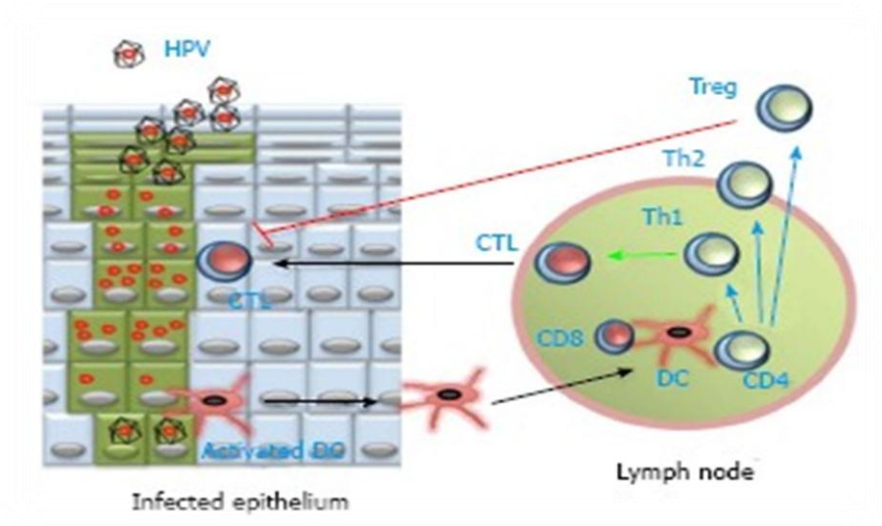


Figure 13 Cellular immune response against human papilloma virus.

Later, antigen presenting cells (APCs), such as dendritic cells (DCs) in the skin and mucosa, Dendritic cells (DC) can take human papilloma virus (HPV) antigens from the infected epithelium, and then migrate (black arrows) to lymph nodes. There, DC present the processed antigen together with MHC class I (HLA in humans) and class II molecules, to CD8+ T cells and CD4+ T cells, respectively. CD4+ T cells proliferate and differentiate (blue arrows) into T helper cells, either Th1 or Th2, depending on the type of cytokines they produce. CD8+ T cells differentiate (blue arrow), with help (green arrow) from Th1 cells, into cytotoxic T cells (CTL). Then, CTL migrate (black arrow) back to the infected epithelium to destroy virus-infected cells. CD4+ T cells can also differentiate (blue arrow) into regulatory T cells (Treg), which inhibit (red line) the cytotoxic activity of CTL [40].

8. Symptoms

In most cases, HPV goes away on its own and does not cause any health problems. But specific types of HPV, it can cause health problems like genital warts and cancer. Genital warts usually appear as a small bump or groups of bumps in the genital area. They can be small or large, raised or flat, or shaped like a cauliflower. A healthcare provider can usually diagnose warts by looking at the genital area. Cervical cancer usually does not have symptoms until it is quite advanced, very serious and hard to treat. For this reason, it is important for women to get regular screening for cervical cancer. Screening tests can find early signs of disease so that problems can be treated early[58], because If untreated, turn into cervical cancer, but usually takes many years." It takes 15 to 20 years for cervical cancer to develop in women with normal immune systems. It can take only 5 to 10 years in women with

weakened immune systems, such as those with untreated HIV infection. When the cervical cancer has reached an advanced stage , many symptoms it appears:

-irregular, intermenstrual (between periods) or abnormal vaginal bleeding after sexual intercourse;

-back, leg or pelvic pain;

-fatigue, weight loss, loss of appetite;

-vaginal discomfort

-a single swollen leg.

Risk factors for HPV persistence and development of cervical cancer

-Early first sexual intercourse

-Multiple sexual partners

-Tobacco use

-Immune suppression (for example, HIV-infected individuals are at higher risk of HPV infection and are infected by a broader range of HPV types) [59]

9. Diagnosis and cytopathic effect

The development of cervical cancer is considered to be A multistep process, where HPV is necessary but in itself an insufficient cause.Disease can only develop when there is persistent HPV infection of the cervical epithelium. Cervical cancer is a rare complication of infection with high-risk HPV, but every abnormal of the cervix is potentially malignant and may develop into cervical cancer over time.Abnormal cervical epithelial cells can be detected microscopically following Papanico-laou (Pap) staining of conventional cervical smears or of the more homogeneous cell suspension from liquid cytology medium. This forms the basis of cervical screening programmes for detection of women at risk of disease progression.

Molecular detection of HPV provides a different approach to screening and patient management. Also, HPV-DNA can be detected in cervical smears and biopsy specimens by various methods, of which in situ hybridization is complementary to cytology, accurate diagnosis of HPV infection relies on the detection of viral nucleic acid. HPV-DNA assays can

be performed using the same specimen as used for cytological examination, which is an important logistic aspect of routine clinical testing. However, a cervical scrape is only a small sample of the cervical epithelium and sampling errors may influence cytology grading. Only a portion of the cervical cell suspension is used for DNA isolation with only a fraction of the isolated DNA being used for specific DNA detection.Therefore, if a specimen only contains a limited number of HPV-DNA copies, sampling errors may produce inconsistencies even in a sensitive assay. This not only has consequences for determining HPV-DNA presence or absence, but also could influence the accuracy of HPV detection, particularly when multiple HPV genotypes are present at different concentrations. However, sampling errors should always be taken into account.Stability of the sample during transport and storage is also important.A wide variety of methods is available for DNA extraction, the choice of which is dependent on origin and quality of the clinical material tested and the diagnostic test used, which should be thoroughly validated for specific laboratory needs [41].

10. Control the virus and prevention

Health education

To protect the community we have To increase health awareness about HPV and other locall y appropriate strategies to promote behaviors that reduce risk of HPV exposure Such behavio rs include delaying onset of sexual activity, reducing the number of sex partners, avoiding sex partners who have multiple partners and using condoms. Who also recommends seeking pro mpt treatment for STI symptoms that may facilitate development of cervical cancer, and avoid ing or reducing tobacco use, which is a known risk factor for cervical cancer.

Early detection through screening.

Cervical cancer prevention programmes should include education (for health-care providers a nd women) that stresses the benefits of screening, the peak ages of cervical cancer incidence, and the signs and symptoms of precancerous lesions and invasive disease . Screening aims to detect precancerous changes, which may lead to cancer if not treated. Screening is only effecti ve if there is a well-organized system for follow-up, diagnosis and treatment. WHO recomme nds specific target ages and frequency of cytologic screening. New programmes should start s creening women at age 30 or older, and should only screen younger women when the older ag e groups have been adequately screened. Existing programmes should not screen women less than 25 years of age. If a woman can be screened only once in her lifetime, the best age is bet

ween 35 and 45 years. For women over 50 years, a five-year screening interval is appropriate. For women aged 25–49 years, a three-year interval can be considered if resources are availabl e. Annual screening is not recommended at any age.

Screening is not necessary for women over 65 years of age, provided the last two previous sm ears in mid-life were negative[1].

Table 1. WHO recommendations for prevention and control of cervical cancer

<table>
<tr><th>Disease</th><th>Primary Prevention</th><th>Early detection and screening</th><th>Treatment</th></tr>
<tr><td rowspan="4">Cervical precancers and cancer</td><td>Reduce high-risk sexual behavior's</td><td rowspan="4">Periodic screening using:
cytology (Pap test) for women aged 25+ years, preferably in organized programmers

In pilot or carefully monitored settings periodic screening with:
HPV DNA tests; or
visual inspection of cervix
with acetic acid or iodine

Prompt diagnostic followup
if screening test abnormal (e.g. colposcopy and biopsy)</td><td>Precancerous lesions: cryotherapy, loop electrosurgical excision procedure, or surgical excision.</td></tr>
<tr><td>Condom use</td><td>“Screen and treat” in low resource settings using cryotherapy</td></tr>
<tr><td>Avoid or reduce tobacco use</td><td rowspan="2">Cancer: surgery, chemotherapy, radiotherapy, brachytherapy, palliative care</td></tr>
<tr><td>Seek prompt treatment of sexually transmitted infections</td></tr>
</table>

11. Treatment

Patients with abnormal screening tests should be referred to colposcopy, the examination of the cervix with a magnification device to visualize precancerous lesions before biopsy or treatment. Although colposcopy is used for cancer screening in some regions [42]. Cryotherapy, loop electrosurgical excision procedure (LEEP) or cold-knife surgical excision can be used to treat precancerous lesions, depending on the location, extent and characteristics of the lesion; the clinician's skills; and the equipment available [60]. Screen-and-treat strategies allow women with positive screening tests to be promptly treated at the same or next clinic visit. This avoids the delays that may mean loss to follow-up or may preclude treatment.

Two approaches:

• Screen-and-treat involves immediate cryotherapy of women with positive screening VIA or VILI at the primary health care facility if lesions are suitable for cryotherapy.

• Screen-triage-treat involves colposcopy of patients with a positive screen (by cytology, VIA, VILI or HPV DNA test) followed by immediate cryotherapy of detected lesions; this approach is intended to minimize the overtreatment associated with the screen-and-treat approach[1]. For example, the Indian study found that a single-visit screen-and-treat approach using VIA, colposcopy and cryotherapy, performed by well-trained nurses, significantly reduced cervical cancer incidence and mortality in women aged 30–59 years [43].

Invasive cervical cancer can be effectively treated with radical surgery, radiotherapy, brachytherapy and chemotherapy, especially if treatment begins when disease is localized to the cervix or uterus 2. Many women in the prime of their lives will suffer prolonged, severe pain; debilitating, embarrassing or offensive symptoms such as bleeding, odorous discharge and abdominal masses; and social isolation from family and friends [42].

11.1 Vaccine

The two currently licensed prophylactic HPV vaccines (bivalent and quadrivalent) are both produced by recombinant technology. By March 2008, both vaccines were licensed in several countries for use in females. Gardasil® is also licensed for males in some countries[1]. Both vaccines are prepared from VLPs produced by recombinant technology , and designed to be prophylactic[44]. The quadrivalent vaccine contains four VLPs – two related to HPV 16 and

18, which are oncogenic, and two for HPV 6 and 11, which are not oncogenic but cause anogenital warts. Produced using common vaccine expression system of a yeast substrate[45]. The bivalent vaccine contains two VLPs – one related to HPV 16 and one related to HPV 18. It produced using a novel baculovirus expression system in Trichoplusian[46]. See the flowing table include more information about both vaccines.

Table 2. Quadrivalent and bivalent HPV vaccine chracteristics

	Quadrivalent vaccine	Bivalent vaccine
Manufacturer and trade name	Merck; Gardasil/Silgard®	GlaxoSmithKline; Cervarix®
Virus-like particles of types	6, 11, 16, 18	16, 18
Substrate	*Saccharomyces cerevisiase* (baker's yeast)	Baculovirus expression system
Adjuvant	Proprietary aluminium hydroxyphosphate sulfate (225 µg) (Merck aluminium adjuvant)	Proprietary aluminium hydroxide (500 µg) plus 50 µg 3-deacylated monophospohoryl lipid A (GSK AS04 adjuvant)
Schedule: 3 doses at intervals of	2 months between doses 1 and 2; 6 months between doses 1 and 3	1 month between doses 1 and 2; 6 months between doses 1 and 3

GSK = GlaxoSmithKline
Source: WHO (2007) (*3*)

11.1.1 Vaccination doses

An illustration of a pragmatic approach quickly translating into impressive reductions in HPV infections and related diseases due to vaccine types is the ongoing school-based vaccination program in Australia. This initiative, which began by temporarily offering free vaccine to females 12–26 years of age, was broadly endorsed and achieved high coverage of both school-aged girls and the older catch-up age group. The 4vHPV vaccine was originally tested and approved as a 3-dose regimen, with a dosing schedule of 0, 2, and 6 months. More recently, a 2-dose schedule (6 or 12 months apart) has been recommended by the World Health Organization for younger age groups (eg, 9–14 years old at first dose), because immunogenicity with 2 doses in preadolescent and early adolescent girls was non inferior to antibody responses in women 16–26 years of age receiving 3 doses [47].

11.1.2 Vaccine mechanism

HPV vaccine was developed as a result of the achievement of core technologies able to produce virus-like particles (VLPs). The recombinant DNA was used to generate VLPs capable of mimicking the natural virus and eliciting high-titers of virus neutralizing antibodies. The L1 gene from the viral genome was sub-cloned in microorganisms, such as yeast (for quadrivalent vaccine) or baculovirus (for bivalent vaccine). In this way L1 over-expressed proteins spontaneously self-assemble into VLPs that resemble the conformation of authentic virions, are neither infectious or nor oncogenic and induce high levels of *type specificneutralizing* antibodies [48].

11.1.3 Side effects

Vaccine as any medicine has side effects . common effects reported after recerved the vaccine include pain, swelling, or redness at the injection site. Fever and nausea also are common side effects . Generally pass within a day or two [49]. US study showed that fainting was not more common after HPV vaccination compared to other vaccines given to teenagers and young women. Therefore, as with other vaccines, a standard fifteen-minute resting period is recommended post-vaccination to prevent any injury associated with fainting[61].

11.1.4 serious effects

There is a lot of warring about both vaccine Gardasil and Cervarix . Events involving hospitalization, death, disability, life threatening illness, or other medically important conditions . Which is extremely rare and no more common than for other vaccines [61].

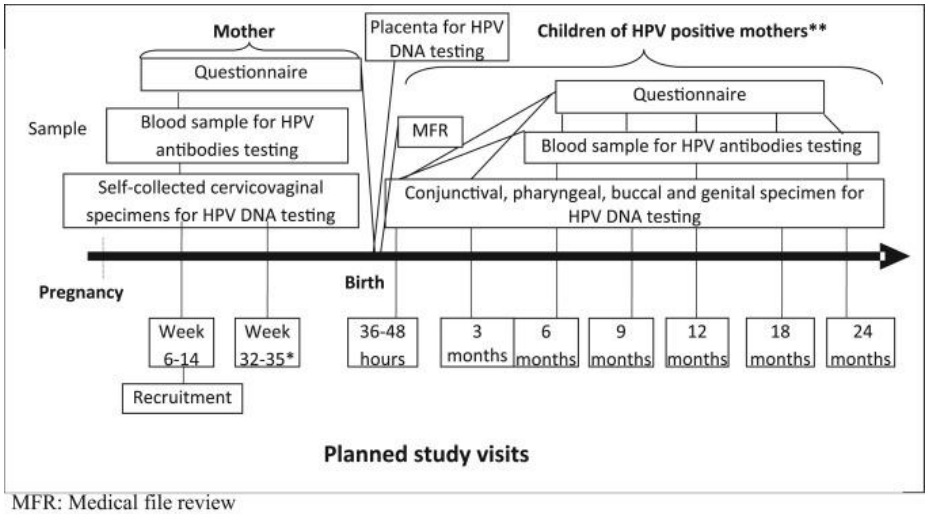
11.1.5 vhp safety

There is a lot of reports involved risk factor and long -term side effect ,but still a few reports can any one write it and published at internet without any strong scientific evidence report of a problem does not mean that the vaccine caused the problem or increased the risk of that event ,only that the event occurred after vaccination. Finally HPV vaccines continue to show very good safety profiles, with no causal links to any deaths and with very low rates of serious side effects[61].Except condition HPV vaccine not recommended for them such as pregnancy,

HIV -positive women and who have had a serious allergic reaction following a specific vaccine or are seriously allergic to anything in a vaccine should discontinue vaccination[61].

12. Recent discoveries

Following research and studies between 2015-2016 concerted about cervical cancer women around world severing and worried . The latest article and research include lots of specific information such as (Molecular archeological evidence in support of the repeated loss of a papillomavirus gene) which study the effect can genome loss while virus replicate, especially E6. the new study looking at losing gene E6 twice [50]. Development of World Health Organization (WHO) recommendations for appropriate clinical trial endpoints for next-generation Human Papillomavirus (HPV) vaccines) The (WHO) serves as a key organization to bring together experts along the continuum of vaccine development and regulatory approval, among its other functions [51]. Other concord study published in 2016 is (Human papillomavirus (HPV) perinatal transmission and risk of HPV persistence among children: Design, methods and preliminary results of the HERITAGE study) the study is very important in my opinion they found that HPV can Perinatal transmission from effective mother to fetal throw placental during pregnancy[52]. The study find very important results such as



MFR: Medical file review
* HPV DNA testing repeated in the third trimester for HPV positive (at 1st trimester) mothers only.
**Only children of HPV DNA positive mother (at 1st trimester) were tested for HPV. <

Figure 14 Design and timeline for information and specimen collection. MFR: Medical file review. *HPV DNA testing repeated in the third trimester for HPV positive (at 1st trimester) mothers only. **Only children of HPV DNA positive mother (at 1st trimester) were tested for HPV.

Found a high prevalence (44.9%) of HPV at prenatal testing in young pregnant women. Furthermore, HPV was frequently detected in placenta samples (14%). There was a non-negligible global prevalence (11.2%) of HPV in newborns (at birth and/or at 3 months of age) from HPV-positive mother. This is also the first study to report transmission of HPV in children's conjunctiva. This interesting data lead to the continuation of the study with an extended phase (phase 2) to increase sample size. The data also seems to be in accordance with recent small studies that report that perinatal transmission (oral and/or genital) is between 4–22% [53-57]. In their meta-analysis published in 2005, Medeiros et al., estimated the perinatal transmission of HPV at 20%. Lots of study recently focus in specific countries population, vaccine effectiveness and side effect. Here a few researches published in 2016 per Elsevier: -

Challenges in educating women about human papillomavirus (HPV) and HPV screening test results: Experience from an HPV demonstration project in North-Eastern Thailand (December 2016)

HPV vaccination intention among male clients of a large STI outpatient clinic in Amsterdam, the Netherlands (December 2016)

Factors affecting HPV vaccine acceptance in west Austria: Do we need to revise the current immunization scheme? (December 2016)

Validation of the vaccine conspiracy beliefs scale (December 2016)

recently, Dr. Ebenezer Tubman's research has been focused on the development of next-generation vaccines against human papillomaviruses (HPVs)—the causative agents of some human cancers (cervical and anogenital cancers). He is exploring strategies to increase immune responses against HPV infections, using platforms such as bacteriophage virus-like particles (VLPs). Dr. Tubman has observed that the display site of an HPV L2 epitope on a bacteriophage VLP platform can affect the magnitude of immune responses to the epitope as well as the breath of protection against HPV infection. He demonstrated that the display of a highly conserved single epitope from HPV L2 protein, on one of the surface-exposed loops of a bacteriophage VLP, induces antibodies that are highly cross-protective against diverse HPV types. Dr. Tubman is a co-inventor of a L2 bacteriophage VLP, which has been licensed to Galva Biotech. He is collaborating with research labs at the University of New Mexico Health Sciences Center as well as a research lab at the National Cancer Institute at the NIH [62].

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