An Insight about Cutaneous Warts: Diagnosis and Immunological Aspects

Hend Darwish Gamil[1], Khaled Mohamed Gharib[1], Kamal Ahmed Mostafa El-Kashishy[2], Amira Mohamed shehata Ibrahim [3,4]

1 Dermatology, Venereology & Andrology Department, Faculty of Medicine, Zagazig University, Egypt

2 Pathology Department, Faculty of Medicine, Zagazig University, Egypt

3 Assistant Lecturer of Dermatology, Venereology & Andrology, Faculty of Medicine, Damietta University, Egypt

4 MSC. of Dermatology, Venereology & Andrology Department, Faculty of Medicine, Zagazig University, Egypt

Corresponding author: Amira Mohamed shehata Ibrahim

E-mail: amiraasker@gmail.com, amiraibrahim@du.edu.eg, amiraaskar@zu.edu.eg
https://orcid.org/0009-0006-0647-0857

Conflict of interest: None declared

Funding: No funding sources

Abstract

Background: While most patients with cutaneous warts are asymptomatic, some may experience physical or psychological discomfort. Cutaneous warts are a common presenting complaint in children and adolescents. Although it is a benign condition, it causes disfigurement, has a tendency to koebnerize, and can be transmitted to others. Common, plantar, or flat warts are cutaneous manifestations of the human papillomavirus. Rapid advances have occurred in the characterization of human papilloma virus (HPV) types applying the new advanced techniques of restriction endonuclease analysis and molecular hybridization to human wart virus. Human papilloma virus can no longer be viewed as a single, homogeneous virus producing all varieties of clinical warts. At least three antigenically heterogeneous HPV types have been associated with common and plantar warts. Two additional HPV types have been found in patients with epidermodysplasia verruciformis. Condylomata acuminata and laryngeal papillomas contain viruses which are also distinct from the preceding viruses and may represent additional HPV types. This antigenic heterogeneity of HPV has important implications concerning the immunology of human warts which have not been taken into account in most previously published studies. Both antibody and cell-mediated responses may be seen in patients with active warts, but many patients with warts have no demonstrable immune reactions. The role of immunity in wart regression remains poorly understood. Nevertheless, the increased frequency of warts in patients receiving immunosuppressive drugs and with immune deficiency states and the immunologic alterations which occur in patients with regressing or cured warts compared to patients with active warts,

particularly the increased frequency of cell-mediated responses and antibodies specific for viral antigens, support a possible role for immunity in the resolution of warts.

Keywords: cutaneous warts, Immunological Aspects

Tob Regul Sci.™ 2021 ;7(6): 6709 - 6718

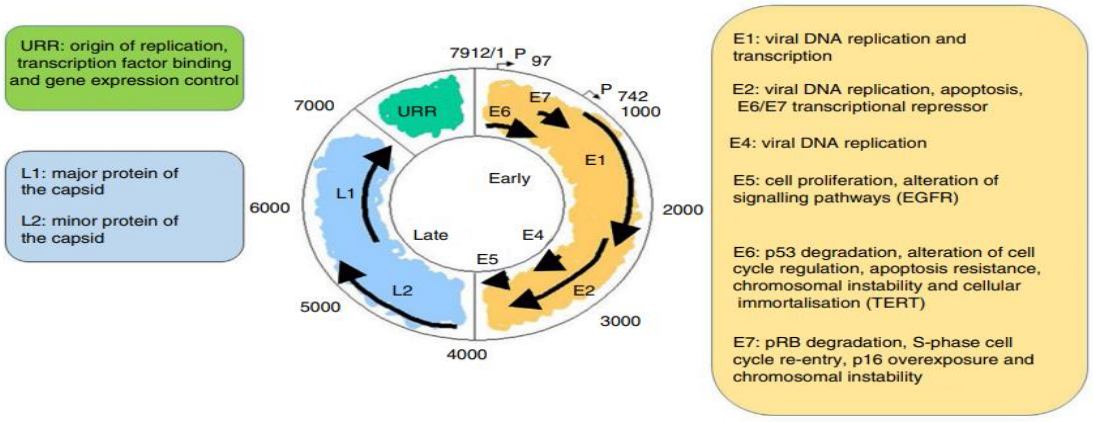
DOI: doi.org/10.18001/TRS.7.6.143

Introduction:

A Skin condition for both patients and practitioners, plantar warts are a source of irritation because no one treatment works for everyone. More than two years of treatment has failed to get rid of recalcitrant warts, thus they're referred to as "recalcitrant." Treating recalcitrant patients with immunotherapy and preventing its recurrence have been good outcomes. HPV is thought to be present in the bodies of 40% of the world's population. Plantar warts affect 14% of the general population each year. When it comes to plantar warts, young people are more likely to have them due to a variety of factors, the most common of which are immunodeficiency or overuse of public showers and/or public restrooms. The majority of cutaneous warts will regress spontaneously within a year or 2 from the initial infection. Reinfection with the same type of HPV virus is uncommon after complete clearance. Transmission occurs via direct skin-to-skin contact or indirectly through contaminated surfaces and objects (e.g., swimming pools, towels, etc.). Minor abrasions and breaks of skin's integrity are usually needed to expose basal keratinocytes to HPV. Autoinoculation of the virus is also a major factor of viral spreading **(1).**

It's common to encounter verrucae, or harmless growths on the skin and mucosa, after infection with the Human papilloma virus [(1)].

Typically, warts are little growths that are rough and hard to the touch and have a similar hue to the rest of the skin. Unless they are located on the soles of the feet, where they can be unpleasant, they rarely cause any symptoms. Hands and feet are the most common sites, although they are not the only ones that might be affected. The number of warts may vary. Because they aren't malignant, they are safe [(2)].



Upstream regulatory region (URR), Telomerase reverse transcriptase (TERT) ,Epidermal growth factor receptor(EGFR).

Figure (1) Structure and function of Human papillomavirus proteins (5).

Prevalence and incidence

There is likely a considerable range in prevalence among different age groups, populations, and time periods (3). Despite the lack of epidemiological data, they are extremely common among youngsters (3). In the general population, the prevalence of warts is stated to be between 1% and 24%. [(4)].

HPV causes warts. In addition, immunosuppression is a risk factor. According to an observational study of immunosuppressed kidney transplant recipients, 90% of those patients had warts five years or more after their donation.[(5)].

Most mucosal HPV types such as HPV6 are also benign, but "high risk" or "oncogenic" genotypes are carcinogenic. While the majority of oncogenic HPV infections are self-limited and subject to immune clearance, persistent infections are associated with a dramatically increased risk for the development of cervical, anogenital and/or oropharanygeal cancers. HPV types 16 and 18 account for 50% and 20% of all cervical cancer cases respectively. Other high/intermediate risk types such as HPV 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 68 constitute the remaining 30% HPV-associated malignancies and are associated with a slower onset of dysplasia **(6)**.

Life cycle of the virus

1) Entry of HPV into the basal cells of stratified epidermis

Human papillomavirus virions invade through damaged areas of the epithelium and infect the basal cells. Although the receptor for the HPV infection has not been fully characterized, the following model has been postulated; virions initially attach to the heparan sulfate proteoglycan (HSPG) on the basal membrane, and transfer to the receptor expressed on the keratinocytes moving on the basal membrane in the wound-healing process, then enter the cells **(7)**.

2) Low-level expression of viral genes and genome maintenance in the basal layer

Following viral entry and uncoating, HPV genomic DNA is transported into the nucleus and maintained at a low-copy number in the basal cells (50 ~ 100 copies per cell; in the basal layer. Genome maintenance as episomal status is essential for the establishment of the early phase of the viral lifecycle **(8)**.

3)Productive replication of HPV in the differentiated cells

After leaving the basal membrane, the infected cells initiate the differentiation program. Because HPV does not encode DNA polymerase activity for viral genome replication, the host DNA replication machinery is required. However, the DNA replication activity is suppressed in the differentiated cells that exit from the cell division cycle. To ensure that the viral genome is replicated, HPV needs to reactivate cell division among the differentiation-initiated cells. E6 and E7 inactivate p53 and retinoblastoma protein (pRb), respectively, which enables the cells to maintain their DNA replication potential. In the upper layers of the stratified epithelium the expression of viral genes that are required for viral genome replication is markedly accelerated, inducing viral genome amplification to thousands of copies per cell. Following the genome

amplification, in the terminally differentiated cells, the synthesis of capsid proteins is triggered. The capsid proteins assemble into virions that encapsidate viral genomic DNA. The progenitor virions are released externally with peeled keratinocytes **(9).**

Human Papillomavirus Evades Innate Immune Defenses

Central to this achievement of immune ignorance is the ability of HPV, particularly the high risk HPV types (HRHPVs), to compromise the role of keratinocytes as innate immune sentinels. Keratinocytes can respond to cell injury and cell stress and can sense pathogens, thus mediating immune responses . Eukaryotic cells express germ line encoded receptors of the innate immune system, pathogen recognition receptors (PRRs) that recognize invariant molecular motifs known as pathogen-associated molecular patterns (PAMPs). There are four groups of PRR: (1) Nucleotide-binding oligomerization domain-like receptors (NLRs), (2) C-type lectin receptors (CLRs), (3) Retinoic acid inducible gene 1 (RIG I)-like receptor family (RLRs), and (4) Toll-like receptors (TLRs) **(9).**

Humoral immunity

B-cells are responsible for humoral response, which neutralize and opsonize viral agents. Humoral immunity is stimulated by antigen presenting cells and Th2 cytokine pattern and depends on CD4 helper T cells that assist B cells to mature and produce antibodies against a specific epitope. The antibodies against HPV target mainly the L1 capsid protein although weak antibodies directing against E2, E6, E7, and L2. All TLRs except TLR3 signal via the MyD88 adaptor protein initiating a cascade of protein protein interactions, and the end result of which is the phosphorylation and activation of IRF7. TLR3 and TLR4 signals via TRIF, the cytosolic RNA sensors signal via MAVS, and the cytosolic DNA sensors via the adaptor protein STING, and the end result of which is the phosphorylation and activation of IRF3. Activated IRF3 and IRF7 homodimerise translocate to the nucleus inducing expression of type I interferons. The HRHPV early proteins, E6, E7, and E2 directly interact with components of these signaling cascades at multiple points, inhibiting signaling via PRR ligation **(10).**

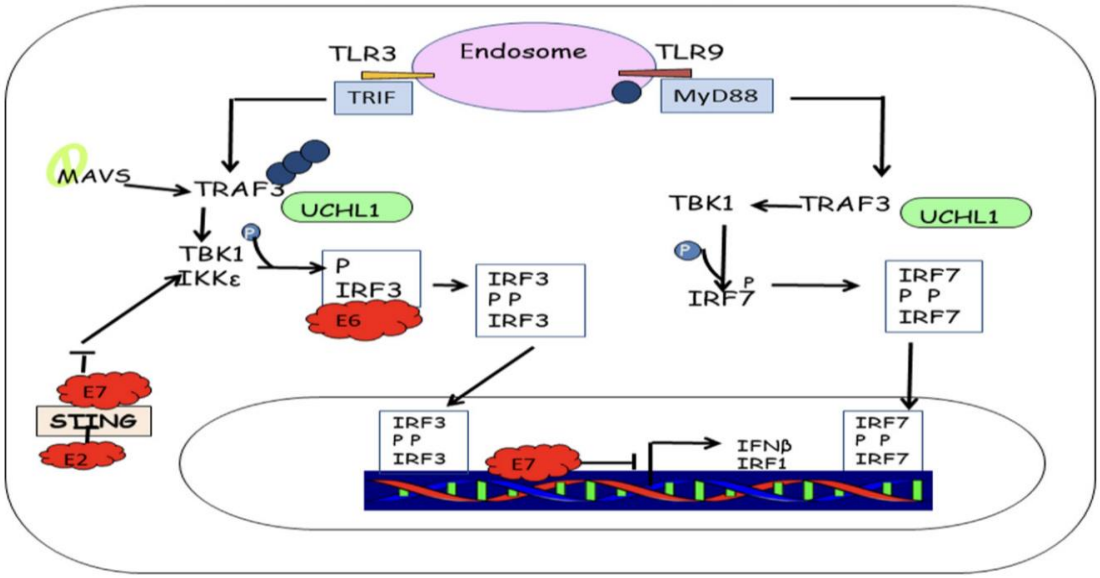


Figure (2) HPV early gene products block signaling from PRRS (10).

Immunity To Human Papillomavirus

HPVs have two central biological properties; they have a tightly restricted host range and exquisite tissue tropism productive virus growth is confined to the fully differentiating squamous epithelia of humans. HPVs are very successful infectious agents. The majority of the β, μ, and ν HPV species reside anonymously in their epidermal niches at very low copy number resulting in no overt disease until, and unless, host immunity is compromised, implying immune control in these scenarios. The α-HPVs, which include most of the types that cause clinical disease, induce chronic infections that have virtually no systemic sequelae, rarely kill the host and, over weeks and months, periodically shed large amounts of infectious virus for transmission to naive individuals (11).

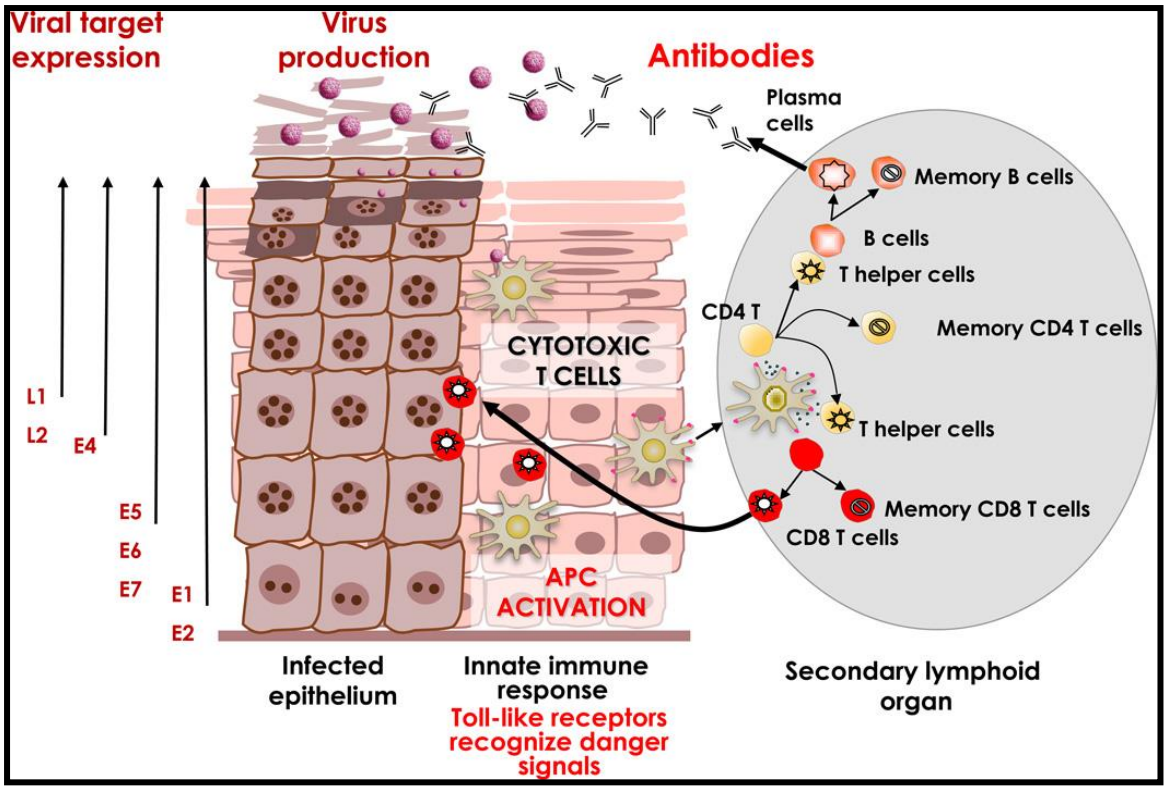


Figure (3) Natural immune control of HPV infection (12).

Diagnosis

The typical clinical appearance of cutaneous warts is typically used to make a diagnosis.

1) Characteristics of the histopathology sample:

Acanthosis, papillomatosis and hyperkeratinization can be seen in a variety of warts, depending on their location. Dermatoscopic examinations reveal reddish or black spots or hairpin or coiled arteries, as well as hemorrhages/extravasation of erythrocytes and elongated and tortuous vessels. (19)

2) Dermoscopy: Vesiculous, yellowish, structureless regions with many irregularly dispersed red-brown to black spots or linear streaks are the hallmarks of palmoplantar warts, which are the result of blood vessel hemorrhages. Palmoplantar warts are characterized by a lack of smooth skin lines. (10)

Studies have shown that HPV27, HPV57, HPV2, HPV1, and HPV4 are the most frequent HPV types detected in samples of common warts worldwide. In immunocompromised individuals skin warts occur more frequently and in greater numbers and are more resistant to treatment. Previous studies have shown that HPV2, HP27, and HPV57 can also be detected in the most common warts of renal transplant recipients and HIV-positive patients, confirming that the distribution of wart-associated HPV types in skin warts is mainly independent of the host's immune system, whereas the diversity of other cutaneous HPV types tends to be higher in skin warts of immunocompromised patients **(11).**

Depending on the clinical appearance, plantar warts can mainly be divided into different subtypes: 1) mosaic plantar warts, which develop as superficially growing and slightly raised clusters of papules with a rough surface, confluent growth, and a characteristic keratinaceous plug surrounded by a hard hyperkeratotic rim, and 2) deep plantar warts (myrmecia), which mainly occur in children between 5 and 15 years old as endophytic growing, deep, smooth-surfaced, single papules at the pressure points on the feet, causing pain especially when walking **(12).**

The most common HPV types detected in plantar warts are HPV57, HPV2, HPV27, HPV1, and HPV4, whereas HPV66 genus, HPV60 and HPV65, and HPV63 were reported with lower frequency. HPV2, HPV27, and HPV57 are commonly detected in mosaic plantar warts, which are notorious for their longevity and resistance to treatment **(6).**

Like common warts, immunocompromised patients have been observed to have a significantly greater number of plantar warts as well as longer duration, higher treatment resistance, and greater frequency of recurrence. HPV2, HP27, and HPV57 can also frequently be detected in plantar warts of HIV-positive patients **(11).**

Warts are usually diagnosed based on their clinical appearance. Plantar warts have special clinical features due to exposure to continuous pressure; they have an endophytic manner, with only a small visible part. In the periungual region, there are often extensive lesions around nails and subungual growth may occur, results in partial onycholysis **(13).**

Common warts were characterized by papillomatosis with prominent parakeratosis at the tips of the digitations, hypergranulosis in the cells between papillations, and mild-to-moderate koilocytic changes in some of the biopsies. Myrmecia also had a papillated contour but, in contrast with common warts, largely exhibited a deeply endophytic profile and striking eosinophilic nuclear and cytoplasmic inclusions in lesional keratinocytes **(13).**

Dermoscopy would be a useful diagnostic tool in cutaneous warts. Papilliform surfaces, interrupted conspicuous skin lines, and homogeneous black to red spots and globules are all regarded as dermoscopic characteristics of viral warts **(14).**

Histopathology is very useful for diagnosis. Acanthosis and hyperkeratosis are observed in viral warts, together with the distinctive koilocytosis observed in higher keratinocytes. There is also papillomatosis in most warts. Basophilic nuclear inclusion bodies, which are observed ultrastructurally to be made of arrays of viral particles, may be present in koilocytes and other granular layer cells. Eosinophilic inclusions resembling uneven, clumped keratohyaline granules are observed in these top epidermal cells **(15).**

Common warts and plantar warts may show thrombosed capillaries, clinically presenting as black dots. These black dots can be visualized by dermoscopy and correspond to the papillary necrosis seen in histology. Histologically, common warts have parakeratosis, acanthosis, and papillomatosis as well as ballooned cells in the spinous layer. Immunohistochemically, HPV can be detected in the granular layer **(16).**

Filiform wart is a digitate (finger-like) collection of tiny fronds that emerges from a face-specific (periorificial), thin pedicle base. Filiform warts are usually found in the beard, nose, and ocular region **(17).**

Flat warts (verruca plana) are typically multiple small flat-topped skin-colored papules most frequently found on the face, hands, and shins. The virus is frequently disseminated by shaving, which results in the spread of warts on the shins and beard area of the face. HPV types 3 and 10 are primarily responsible for plane warts **(8).**

Butcher's warts have a cauliflower-like appearance and are specifically brought on by HPV 7 infection in the hands of butchers and other workers whose skin is chronically macerated due to moisture and cold **(18).**

Pigmented warts (HPV types 4/60/65), petechial warts (HPV type 63), ridged warts (HPV type 60), white small warts/small wart-like (HPV types 88/95), and viral plantar epidermal cysts (HPV types 57/60) are also reported **(19).**

Epidermodysplasia Verruciformis (EV) is an autosomal recessive disorder caused by mutations in two genes of the Transmembrane Channel-like Protein Complex family (TMC6 and TMC8) encoding two transmembrane proteins that form a complex with intracellular Zinc transporter proteins. Affecting its distribution and downregulating zinc-associate transcription factors that cause discrete flat-topped papules resembling flat warts, flat pityriasis versicolor-like lesions and high risk of squamous cell carcinoma **(20).**

Anogenital warts (Condylomata acuminate) can spread to the nearby inguinal folds, perineum, and perianal region. Typically, they take the form of fleshy, elevated papules that range in size from one to five millimeters. They can be broad and flat, pedicled, occasionally resembling a cauliflower, or huge tumor masses called Buschke-Lowenstein tumors that take up the entire anogenital region. HPV strains 6 and 11 are the most prevalent strains that cause condyloma acuminata. High-risk subtypes of HPV types 16 and 18 increase the risk of acquiring cancer **(21).**

Bowenoid papulosis is a sexually transmitted disorder thought to be caused by human papillomavirus type 16. They appear reddish brown or violet in color, small, solid, smooth, raised, and velvety most commonly involves the anogenital area, but extragenital disease is reported and may present on other parts of the genitals, around the anus, or oral cavity. It is usually asymptomatic, but lesions can become inflamed, itchy, and painful **(22).**

Oral warts are small soft pink papules that appear on the buccal, labial, gingival or palatal mucosae caused by HPV6 and 11 genotypes. in AIDS patients, HPV types 7, 71, 72 and 73 were Unusually found to be associated with them **(23). Focal epithelial hyperplasia or Heck's disease** is a rare, benign, oral condition that is associated with infection by human papillomavirus type 13, 32 or

both. Normal asymptomatic white to mucosal-colored, soft, papular or nodular raised lesions in the oral cavity might expand to a size or location requiring treatment **(24).**

Immunological Aspects of Warts

Human papillomavirus (HPV) is a nonenveloped double stranded circular DNA virus consisting of E1, E2, E4, E5, E6, E7, L1, and L2 genes with strains capable of causing cutaneous warts, as well as cancers of the cervix, vagina, vulva, anus, penis, oral cavity, and oropharynx **(25).**

The productive life cycle of HPV can be loosely grouped into 3 phases: establishment, maintenance, and productive amplification**.** The establishment phase involves viral transcription and genome amplification following nuclear entry. The viral proteins E1 and E2 are involved in viral genome amplification and viral DNA is initially maintained as episomes where viral gene expression is tightly regulated. E2 is a DNA binding protein and plays an important role in the initiation of viral DNA replication. Ultimately, each host cell will contain approximately 50-100 viral episome **(26).**

The maintenance phase is initiated to uphold a constant number of viral genomes and to establish a persistent infection. E2 tethers the viral genome to host chromosomes and supporting viral genome during mitosis. HPV genomes can be maintained in basal epithelial cells for years to several decades. Less than 10% of new infections lead to persistent infections, dysplasia and cancer implying a role for immune mediated clearance of virally infected cells. **(27).**

At some point, HPVs switch from stable maintenance of the viral genome to productive replication which occurs in the differentiating epithelial cells. Normally, differentiating cells would not be capable of supporting viral DNA synthesis, however, the viral proteins E6 and E7 activate host cell DNA replication machinery allowing for continued viral DNA synthesis **(28).**

HPV DNA can integrate into the host DNA over the long maintenance phase, and this is commonly found in both premalignant and malignant lesions. Integration usually disrupts the E1 and E2 genes, which terminates E2 driven transcriptional repression of E6 and E7 leading to increased oncogene expression **(29).** Viral integration is inversely correlated with circulating viral load (integrated virus cannot replicate and release new virions) **(30)**

References:

1. Sterling JC, Handfield-Jones S and Hudson PM (2014): Guidelines for the management of cutaneous warts. British Journal of Dermatology, 144(1), 4-11.
2. Rollins MD and Vanderhooft SL (2017): Benign Skin Lesions. In Fundamentals of Pediatric Surgery. Springer, Cham, 853-862.
3. Lacarrubba F, Verzì AE and Quattrocchi E (2018): Cutaneous and Anogenital Warts. Atlas of Pediatric Dermatoscopy. Springer, 35-44.
4. Chen GY, Cheng YW and Wang CY (2008): Prevalence of skin diseases among schoolchildren in Magong, Penghu, Taiwan: a community-based clinical survey. Journal of the Formosan Medical Association 107: 21-29.
5. Reusser, N. M., Downing, C., Guidry, J., & Tyring, S. K. (2015): HPV carcinomas in immunocompromised patients. Journal of clinical medicine, 4(2), 260-281.

6. de Planell-Mas E, Martínez-Garriga B, Zalacain AJ (2017): Human papillomaviruses genotyping in plantar warts. Journal of Medical Virology; 89: 902-907.
7. Al Aboud AM and Nigam PK (2017): Wart (Plantar, Verruca Vulgaris, Verrucae). StatPearls. StatPearls Publishing.
8. Bruggink SC, Eekhof JA and Egberts PF (2013): Warts transmitted in families and schools: a prospective cohort. Pediatrics: PEDS. 2012-2946.
9. Park, I. U., Introcaso, C., & Dunne, E. F. (2015): Human papillomavirus and genital warts: a review of the evidence for the 2015 centers for disease control and prevention sexually transmitted diseases treatment guidelines. Clinical Infectious Diseases, 61(suppl_8), S849-S855.
10. De Koning M, Quint K and Bruggink S (2015): High prevalence of cutaneous warts in elementary school children and the ubiquitous presence of wart associated human papillomavirus on clinically normal skin. British Journal of Dermatology; 172: 196-201.
11. Skubic Nuovo G, Nicol A, de Andrade CV, et al. (2019a): New biomarkers of human papillomavirus infection in epidermodysplasia verruciformis. Ann Diagn Pathol.40; 81-87.
12. Witchey DJ, Witchey NB, Roth-Kauffman MM, et al. (2018): Plantar warts: epidemiology, pathophysiology, and clinical management. JOM.118; 92-105.
13. Abeck D, Tetsch L, Lüftl M, et al. (2019): Extragenital cutaneous warts–clinical presentation, diagnosis and treatment. J Dtsch Dermatol Ges.17; 613-634.
14. Al Rudaisat Ma & Cheng H (2021): Dermoscopy features of cutaneous warts. Int J Gen Med.16; 9903-9912.
15. Chauhan K, Jassal V, Sara GK, et al. (2021): Histopathological study of verrucous lesions and its mimics. J Microsc Ultrastruct.9; 86-97.
16. Alaziz M & Najim N (2021): Histological differential diagnosis of verruca vulgaris: A systematic review. Int J Clin Diagn Pathol.4; 26-31.
17. Strumia R (2016): Verruca Filiformis (Filiform Wart). In: Abramovits, W, Graham, G, Har-Shai, Y, et al. (eds.) Dermatological Cryosurgery and Cryotherapy. London: Springer. 631-632.
18. Miyata K, Go U & Mitsuishi T (2021): So-Called Butcher's Warts Appeared on the Hands of a Meat Handler. Case Rep Dermatol.12; 219-224.
19. Shimizu A, Yamaguchi R & Kuriyama Y (2023): Recent advances in cutaneous HPV infection. J Dermatol.50; 290-298.
20. Ün M, Durmaz OB & Köylüoğlu E (2020): Epidermodysplasia verruciformis. J Exp Med Sci.1; 042-045.
21. Lacey CJ, Guimera N & Garland SM (2020): Low-risk Human Papillomavirus: Genital Warts, Cancer and Respiratory Papillomatosis. Human Papillomavirus. Elsevier. 165-178.
22. Seong SH, Jung JH, Kwon DI, et al. (2021): Dermoscopic findings of genital keratotic lesions: Bowenoid papulosis, seborrheic keratosis, and condyloma acuminatum. Photodiagnosis Photodyn Ther.36; 102448.
23. Di Spirito F, Pantaleo G, Di Palo MP, et al. (2023): Oral human papillomavirus benign lesions and HPV-related cancer in healthy children: a systematic review. Cancers (Basel).15; 1096.
24. Bendtsen SK, Jakobsen KK, Carlander A-LF, et al. (2021): Focal epithelial hyperplasia. Viruses.13; 1529.
25. Ilyas S (2022): Human Papillomavirus: Detection Method and Infection. IJoEP.4; 82-91.

26. Burley M, Roberts S & Parish JL (2020): Epigenetic regulation of human papillomavirus transcription in the productive virus life cycle. Semin Immunopathol.42; 159-171.
27. Pim D, Broniarczyk J, Siddiqa A, et al. (2021): Human papillomavirus 16 L2 recruits both retromer and retriever complexes during retrograde trafficking of the viral genome to the cell nucleus. J Virol.95; 10.1128/jvi. 02068-20.
28. Ozbun MA & Campos SK (2021): The long and winding road: Human papillomavirus entry and subcellular trafficking. Curr Opin Virol.50; 76-86.
29. Bhat D (2022): The 'Why and How'of cervical cancers and genital HPV infection. Cytojournal.19.
30. Letafati A, Sakhavarz T, Khosravinia MM, et al. (2023): Exploring the correlation between progression of human papillomavirus infection towards carcinogenesis and nutrition. Microb Pathog.106302.