Mechanisms of Viral Infections Associated with HIV: Workshop 2B

S.M. Tugizov[1]*, J.Y. Webster-Cyriaque[2], S. Syrianen[3], A. Chattopadyay[4], H. Sroussi[5], L. Zhang[6], and A. Kaushal[7]

[1]Department of Medicine, School of Medicine, University of California, San Francisco, USA; [2]School of Dentistry, University of North Carolina at Chapel Hill, North Carolina, USA; [3]Institute of Dentistry and MediCity Research Laboratory, Faculty of Medicine, University of Turku, Turku, Finland; [4]National Institutes of Health, National Institute of Dental and Craniofacial Research, Bethesda, Maryland, USA; [5]University of Illinois at Chicago, College of Dentistry, Chicago, Illinois, USA; [6]State Key Laboratory of Bioorganic and Natural Products Chemistry, Shanghai Institute of Organic Chemistry, Chinese Academy of Sciences, Shanghai, China; and [7]National Center for Safety Pharmacology, Department of Pharmacology and Toxicology, National Institute of Pharmaceutical Education and Research, Punjab, India; *corresponding author, sharof.tugizov@ucsf.edu

Adv Dent Res 23(1):130-136, 2011

ABSTRACT

HIV infection is commonly associated with activation and dissemination of several other viral pathogens, including herpes simplex virus 1/2, human cytomegalovirus, human herpesvirus 8, Epstein-Barr virus, Varicella Zoster virus, and human papillomavirus, which behave as opportunistic agents and cause various diseases in immunocompromised hosts. The increased frequency and severity of diseases caused by these viruses in HIV-infected individuals is due mainly to dysfunction of both the adaptive and innate immune responses to viral pathogens. In addition, molecular interactions between HIV and these opportunistic viruses are likely to play critical roles in the progression of disease, including neoplasia. This report reviews the critical aspects of HIV interaction with opportunistic viruses, including Epstein-Barr virus, human cytomegalovirus, herpes simplex virus, Varicella Zoster virus, human herpesvirus 8, and human papillomavirus.

HIV infection and the development of AIDS lead to opportunistic infection by several common viruses, such as herpes simplex virus 1/2 (HSV-1/2), human cytomegalovirus (HCMV), Epstein-Barr virus (EBV), human herpesvirus 8 (HHV-8), Varicella Zoster virus (VZV), and human papillomavirus (HPV) (Mbopi-Kéou *et al*., 2002; Hagensee *et al*., 2004; Leigh *et al*., 2004; Palefsky, 2009). Furthermore, HIV infection leads to asubstantial depletion of $CD4^+$ T cells in peripheral blood, lymphoid organs, and mucosal tissues, causing $CD8^+$ T-cell dysfunction (Feller *et al*., 2008; Levy, 2009). $CD4^+/CD8^+$ T cells play critical roles in the immune defense against almost all human pathogens, including viruses, and the depletion and dysfunction of these immune cells in HIV-positive patients can lead to the activation of herpesviruses (Griffin *et al*., 2008; Carbone *et al*., 2009), which are usually only latent under normal immune surveillance (Roizman and Whitley, 1993). Immune dysfunction caused by HIV may also promote infection and/or reactivation of HPVs, including the high-risk HPV types HPV 16 and HPV 18 (Hagensee *et al*., 2004; Leigh *et al*., 2004; Palefsky, 2009). HIV-associated reactivation of opportunistic viruses is known to cause several clinically important diseases, including malignant and nonmalignant lesions of the oral epithelium (Hagensee *et al*., 2004; Palefsky, 2009). Although highly active antiretroviral therapy (HAART) is efficient in inhibiting HIV replication and recovery of the immune system, it does not substantially reduce the mucosal shedding of herpesviruses (Griffin *et al*., 2008). Furthermore, HAART does not reduce the prevalence of HPV infection in mucosal epithelia (Hagensee *et al*., 2004; Palefsky, 2009).

In this report, we review the evidence for molecular interactions between HIV and various opportunistic viruses, as well as the role of such interactions in viral activation and spread. We also discuss the potential role of HAART in HPV infection and activation and in the development of HIV/AIDS-associated malignancies. As such, we address the following questions:

Question 1: How does HIV interact with the oral opportunistic pathogens EBV, HCMV, HSV, VZV, HPV, and HHV-8?
Question 2: Does HIV alter HSV-1 transmission, and vice versa?
Question 3: What oral malignancies are associated with HIV/AIDS? Which is primary and which is secondary—does one predispose an individual to the other?
Question 4: How does HAART relate to oral HPV infection and disease, warts, and cancer?
Question 5: What immune mechanisms make HIV+ patients susceptible to other viral infections?

QUESTION 1

How does HIV interact with the oral opportunistic pathogens EBV, HCMV, HSV, VZV, HPV, and HHV-8?

Key Words

HIV/AIDS, infectious disease, treatment planning, HSV-1 transmission, HAART, oral malignancy.

DOI: 10.1177/0022034511400076

HIV-associated activation and reactivation of opportunistic viruses in the oral cavity creates a favorable microenvironment for direct and indirect interaction between HIV and opportunistic viral pathogens. Cell-free HIV-1 virions and viral DNA/RNA can be isolated from saliva and oral mucosal epithelium of HIV/AIDS patients (Kakizawa *et al*., 1996; Qureshi *et al*., 1997; Chou *et al*., 2000; Maticic *et al*., 2000). HIV-infected lymphocytes and macrophages can be detected in the mucosal and submucosal layers of oropharyngeal epithelium (Chou *et al*., 2000; Jayakumar *et al*., 2005; Rodriguez-Inigo *et al*., 2005), and electron microscopy can detect HIV virions within the tight junctions of oral epithelium (Qureshi *et al*., 1997). Analysis of integrated HIV-proviral DNA by *in situ* polymerase chain reaction assay has shown that the HIV genome is present within the basal layer of stratified buccal epithelium (Qureshi *et al*., 1997) and in Langerhans cells within mucosal and submucosal layers of hairy leukoplakia lesions (Chou *et al*., 2000). Presence of HSV-1, HCMV, EBV, HHV-8, VZV, and HPV in the oral lesional epithelium and saliva is well documented (Greenspan *et al*., 1985; McCullough and Savage, 2005; Miller *et al*., 2006; Tugizov *et al*., 2007). Thus, HIV and opportunistic viruses may replicate in the mucosal intraepithelial and subepithelial lymphocytes, macrophages, and Langerhans cells and in mucosal epithelial cells, and such close coexistence of these pathogens in the oral mucosal compartment may increase their direct or indirect interactions. Furthermore, shedding of HIV and opportunistic viruses into saliva may allow their direct interaction with the mucosal surface. Interaction of viral proteins with mucosal epithelial cells, as well as intraepithelial and subepithelial lymphocytes, macrophages, and Langerhans/dendritic cells, may modulate multiple signaling events at the cellular and tissue levels and therefore play a significant role in the pathogenesis of HIV-associated oral mucosal disease.

One of the best-known signaling pathways modulated by viruses is the nuclear factor kappa–light chain enhancer of activated B cells (NF-κB), which is involved in the regulation of transcription of viral and cellular genes. HIV tat protein activates phosphoinositide 3-kinase (Ganju *et al*., 1998; Milani *et al*., 1998; Urbinati *et al*., 2005), which in turn induces IκB kinase, leading to the translocation of NF-κB to nuclei. HIV tat is efficiently secreted from HIV-infected cells and internalized by nearby cells (Ensoli *et al*., 1993; Chang *et al*., 1997). Tat protein exits from cells via a Golgi-independent, leaderless secretory pathway (Chang *et al*., 1997). Tat's basic domain (49-57 aa) contains a unique motif (RKKKRRQRRR) called the protein transduction domain, which is responsible for its penetration/endocytosis into cells and subcellular localization (Vives *et al*., 1997; Nagahara *et al*., 1998). Tat's protein transduction domain may mediate its spread within tissues using endocytosis and transcellular transcytosis pathways. For example, within a few hours after intraperitoneal injection of a β-galactosidase fusion protein containing the tat protein transduction domain into mice, β-galactosidase was detected in all tissues, including liver, kidney, heart, lung, and brain (Schwarze and Dowdy, 2000). Peptides fused to the tat-BD domain have been shown to migrate across polarized epithelial cells by endocytosis and transcytosis (Lindgren *et al*., 2004; Herve *et al*., 2008). HIV tat has also been shown to penetrate the superficial part of the intact stratified epithelium of the cornea and wounded cornea, allowing rapid and intensive penetration of tat into the deeper stratified epithelium and stromal tissue (Guo *et al*., 2004). Thus, secretion of HIV-tat from intraepithelial and subepithelial immune cells may lead to its penetration into mucosal epithelial cells, where it can activate NF-κB. HIV-tat-mediated activation of NF-κB in epithelial cells may activate latent herpesviruses, because most herpesviral promoters have response elements to NF-κB (Santoro *et al*., 2003). Activation of NF-κB may also lead to the upregulation of HPV oncoproteins, facilitating the progression of HPV neoplastic processes (Ruutu *et al*., 2002; Kim *et al*., 2009). HIV-tat also activates HPV replication, its protein expression, and the proliferation of HPV-infected keratinocytes (Vernon *et al*., 1993; Buonaguro *et al*., 1994; Kim *et al*., 2008). These findings suggest that HIV tat may activate gene expression and/or replication of opportunistic viruses within the oral mucosa and contribute to the development of HIV-associated oral diseases.

HIV-associated activation and dissemination of opportunistic viruses within the oral mucosal epithelium may occur by HIV-mediated activation of proinflammatory cytokines. Activation and secretion of proinflammatory cytokines—such as IL-12, IL-6, IL-8, and TNF-α—in HIV-infected intraepithelial macrophages and dendritic cells induces apoptotic and nonapoptotic changes in intestinal epithelial cells and disrupts their barrier function (Stockmann *et al*., 2000; Schmitz *et al*., 2002). HIV-associated barrier dysfunction may also attenuate the innate immune functions of the intestinal epithelium (Sankaran *et al*., 2008). The functional effects of barrier and innate immune impairment of the oral epithelium are not yet known in the context of HIV-mediated upregulation of proinflammatory cytokines. Disruption of the oral epithelial barrier may lead to the opening of paracellular spaces and the reentry of HSV-1/2, HCMV, EBV, VZV, or HPV, which can all be secreted at high levels into saliva (Fidouh-Houhou *et al*., 2001; Brengel-Pesce *et al*., 2002; Miller *et al*., 2006). Furthermore, disruption of mucosal epithelia may allow superinfection by new strains of these viruses. For example, disruption of the corneal epithelium was found to lead to superinfection by HSV-1 (Remeijer *et al*., 2002). Similarly, disruption of the intestinal mucosal epithelium in inflammatory bowel disease causes superinfection by HCMV (Berk *et al*., 1985). Thus, HIV-associated disruption of the oral epithelium may lead to penetration by HSV-1/2, HCMV, EBV, VZV, or HPV via paracellular spaces, allowing them to infect the immune and epithelial cells of the oral mucosa and leading to local and systemic opportunistic infection.

Opportunistic viruses may also activate HIV replication within the oral microenvironment by interacting with the HIV transcription machinery. Herpesviral immediate early proteins transactivate the HIV long terminal repeat (LTR) and therefore facilitate HIV replication. HSV-1 immediate early proteins ICP0, ICP4, and ICP27 and transactivating proteins IE110 and IE175 all induce the transactivation of HIV-1 LTR by upregulating the NF-κB and SP1 transcription factors (Mosca *et al*., 1987; Margolis *et al*., 1992). Similarly, HCMV immediate early proteins IE1 and IE2 promote the transactivation of the HIV-1 LTR

by upregulating NF-κB (Ghazal *et al*., 1991; Yurochko *et al*., 1995). The EBV nuclear antigen EBNA-2 may also transactivate the HIV LTR in synergy with HIV tat (Zhang *et al*., 1997). Finally, HIV LTR has been shown to be activated by HHV-8 immediate early protein KIE2 and latency-associated nuclear antigen (Hyun *et al*., 2001).

These findings suggest that in the oral cavity, HIV may directly and/or indirectly interact with HSV-1/2, HCMV, EBV, VZV, and HPV and that these interactions may have synergistic effects on the replication of both HIV and opportunistic viruses, promoting the progression of viral disease within the oral mucosa.

QUESTION 2

Does HIV alter HSV-1 transmission, and vice versa?

HIV-associated HSV-1 activation is a common oral manifestation of HIV/AIDS disease. One of the critical mechanisms of HSV-1 activation in HIV-infected immunocompromised individuals is HIV-mediated immune dysfunction (Griffin *et al*., 2008). In healthy individuals, HSV infection is mainly latent but can be reactivated by various stimuli, including physical and emotional stress, after which it usually spreads only locally (Roizman and Whitley, 1993; Lachmann, 2003). Both the humoral and cellular immune systems are critical for control of HSV replication upon its reactivation (Corey *et al*., 2004; Griffin *et al*., 2008). The neutralizing antibodies against herpesviral glycoproteins play a critical role in controlling of viral reactivation (Corey *et al*., 1999; Mott *et al*., 2007). The NK cells and cytotoxic T lymphocytes may play a major role in elimination of HSV-1 infected cells (Koelle *et al*., 1998; Reading *et al*., 2006). HIV-associated immune deficiencies, including $CD4^+$ T-cell depletion and $CD8^+$ T-cell dysfunction, lead to impairment of immune control of HSV replication and may therefore cause development of extensive mucocutaneous lesions (Griffin *et al*., 2008). HIV-associated extended HSV-1 reactivation and development of chronic lesions may result in extensive shedding of infectious virions into saliva, which may serve as a potential source of HSV for transmission to other individuals.

Although HIV transmission through the adult oral epithelium is rare (Tudor-Williams and Lyall, 1999; Page-Shafer *et al*., 2006), HSV-1-associated oral lesions may facilitate HIV transmission across this epithelium because of disruption of epithelial integrity. Furthermore, in HIV-infected individuals, HSV-induced disruption of the oral epithelium may serve as a portal for the entry of new HIV strains. Moreover, HIV transmission might be further facilitated by migration of HIV-susceptible immune cells toward the disrupted epithelium as a result of the inflammatory processes associated with herpetic lesions (Corey, 2007; Rebbapragada *et al*., 2007). HIV and HCMV (Maheshwari *et al*., 2006; Fox-Canale *et al*., 2007), as well as HIV and HSV-1, can coinfect the same macrophages or epithelial cells *in vivo* (Heng *et al*., 1994). Thus, within the herpetic lesions, some HIV-susceptible cells might be infected with HIV and HSV-1. As we describe above, this could lead to upregulation of HIV replication via transactivation of its LTR by HSV immediate early proteins and transactivating proteins.

QUESTION 3

What oral malignancies are associated with HIV/ AIDS? Which is primary and secondary—does one predispose to the other?

Accumulated evidence indicates that HIV/AIDS facilitates development of neoplastic processes, including oral malignancies such as Kaposi sarcoma, B-cell lymphomas (Burkitt lymphoma, Hodgkin disease, non-Hodgkin and plasmablastic lymphomas), and basal and squamous cell carcinomas (Epstein, 2007; Carbone *et al*., 2009). The incidence of these cancers in HIV-infected patients is about 2- to 3-fold higher than that in HIV-negative individuals. Kaposi sarcoma is the most common malignancy in HIV/AIDS. Oral Kaposi sarcoma is an HHV-8-infected and angioproliferative malignant lesion, which can appear in the palate, gingiva, and tongue (Carbone *et al*., 2009). Interestingly in the HAART era, Kaposi sarcoma even persists in patients with undetectable HIV viral loads and CD4 counts above 300 (Mani *et al*., 2009). HIV-associated B-cell lymphomas mostly originate from EBV-infected B-cell lymphoblasts, and they may develop into soft tissue masses within the gingival and palate mucosa and the mandible (Carbone *et al*., 2009). The plasmablastic lymphoma of the oral cavity also originates from lymphoproliferative B cells, which are frequently infected with EBV; however, the plasmablastic lymphoma cells do not express B-cell markers (CD20 and PAX5) but, rather, the plasma cell markers (CD138/syndican-1, MUMI/IRF4, and VS38c), indicating that these cells may have an unusual immunophenotype (Carbone *et al*., 2009). Although introduction of HIV HAART has substantially reduced HIV replication and HIV-mediated immune system dysfunction, it has not affected the incidence of HIV/AIDS-associated lymphomas—particularly, non-Hodgkin lymphomas (Epstein, 2007).

Oral squamous cell carcinomas in HIV-infected individuals have been detected in the oral and oropharyngeal mucosal epithelium and lips, and these cancers are typically found at more advanced stages and have poorer survival rates (Epstein, 2007). Malignant and nonmalignant oral squamous lesions and warts are mostly associated with high-risk HPV types 16, 18, and 31 (Hagensee *et al*., 2004). Furthermore, tobacco and alcohol use may serve as risk factors for development of oral squamous cell carcinomas (Epstein, 2007).

As described above, most HIV/AIDS-associated oral cancer cells are infected with HHV-8, EBV, or HPV, suggesting that loss of cytotoxic T-lymphocyte responses or direct and indirect molecular interactions between HIV and these opportunistic viruses may lead to the promotion of neoplastic processes. Synergistic activation of HIV, EBV, HHV-8, and HPV in oral mucosa may enhance the oncogenic role of EBV, HHV-8, and HPV and may therefore increase the incidence of oral neoplastic processes. NF-κB-dependent cytokines—including TNF-α, IL-1, IL-6, and IL-8—in the saliva of patients with malignant and premalignant lesions are elevated (Rhodus *et al*., 2005), suggesting that abnormal upregulation of the these proinflammatory cytokines may promote the development of oral malignancies. Because HIV and oral opportunistic virions may activate these proinflammatory cytokines in oral mucosa, virus-mediated cytokine induction may facilitate oral neoplasia.

HIV/AIDS-associated primary cancer may predispose and initiate secondary cancers. For example, in a case study, 2 HIV-infected patients first developed HPV-related anal squamous cell carcinoma (Chaiyachati *et al*., 2008), then later developed oral squamous cell carcinoma. These events suggest that anal-genital and oral-genital sexual contacts facilitate dissemination of high-risk HPVs from the anal site to the oral site and therefore initiate subsequent neoplastic process from anal mucosa into oral mucosa, or vice versa.

QUESTION 4

How does HAART relate to oral HPV infection and disease, warts, and cancer?

HIV infection is consistently associated with a higher prevalence and incidence of HPV infection, which is well correlated with the development of benign and malignant mucosal epithelial lesions, including those in the oral epithelium (Hagensee *et al*., 2004; Leigh *et al*., 2004; Palefsky, 2009). Although HIV HAART drastically reduces oral lesions associated with HSV-1 and HCMV and corresponds with a significant decline in the EBV-associated hairy leukoplakia lesion (Patton, 2000), it does not have a significant therapeutic effect on the development of HPV-infected malignant or nonmalignant lesions (Hagensee *et al*., 2004; Hodgson *et al*., 2006; Cameron and Hagensee, 2008; Palefsky, 2009). HIV HAART-associated increase of HPV infection has been detected with high-risk HPV types 16, 18, and 31, which can cause malignant and nonmalignant lesions of oral mucosal epithelium (Hagensee *et al*., 2004). Overall, the prevalence of HPV infection or precancer in those on HAART has not diminished.

The mechanisms of HAART-associated increase in HPV infection are not well understood. The increase could be due to the natural history of HPV infection—that is, from the initial entry of virus into the basal epithelium to the full manifestation of malignant/nonmalignant epithelial lesions. It is possible that, before the HAART era, HIV/AIDS patients died before the appearance of HPV-infected lesions and that HAART-associated prolongation of life in HIV-infected patients now allows the development of HPV-mediated benign and neoplastic changes. HAART-mediated immune reconstitution might also be associated with the appearance HPV-infected lesions owing to upregulation of proinflammatory chemokine/cytokines, which may facilitate activation of HPV replication (Meys *et al*., 2010). However, an analysis of epithelial cells of HPV-infected warts did not indicate significant upregulation of chemokine/cytokines (Lilly *et al*., 2005), suggesting that secretion of chemokine/cytokines from activated immune cells may promote HPV replication in a trans manner.

The increase in HIV HAART-associated HPV infection may be related to HAART treatment. As such, HAART may affect the key functions of mucosal epithelia, including their differentiation and stratification, and may therefore reduce the barrier function of the oral epithelium, leading to penetration of HPV to the basal cells—that is, disruption of the oral epithelium results in a higher level of HPV infection. It is also possible that HAART modulates gene expression in epithelial cells and so may increase the binding, entry, and/or replication of HPV. Finally, components of HAART may directly interact with HPV proteins and activate HPV replication and/or gene expression.

QUESTION 5

What immune mechanisms make HIV+ patients susceptible to other viral infections?

The immune mechanisms of HIV-associated susceptibility of opportunistic viruses—including HSV-1/2, HCMV, EBV, VZV, and HPV—is mainly due to HIV-mediated impairment of systemic and local (*i.e.*, mucosal) immune systems (Leigh *et al*., 2004; Paiardini *et al*., 2008). It was recently determined that HIV-mediated suppression of interferon regulatory factor 3 promotes HIV-1 infection and disables the innate antiviral defenses of the host cell. Global disruption of innate antiviral responses by HIV-1 may increase susceptibility of host cells to various opportunistic viral infections (Doehle *et al*., 2009). HIV infection and depletion of $CD4^+$ T cells leads to dysfunction of $CD4^+$ T lymphocytes, which is a key player in the control of opportunistic viruses. HIV-associated dysfunction of the T-lymphocyte-based systemic immune system results in the weakening or lack of surveillance mechanisms for reactivated opportunistic viruses and therefore leads to their uncontrolled replication. Subsequently, HIV-associated impairment of the mucosal immune system promotes spread of opportunistic viruses within the mucosal epithelium. Although HIV-caused systemic immune dysfunction has been investigated in great detail, the HIV-associated immune dysfunction of the mucosal epithelium—particularly, the oral mucosal epithelium—has not been well investigated. Analysis of the gut mucosal epithelium has shown that HIV infection may cause rapid and substantial depletion of mucosa-associated lymphoid tissue–based memory $CD4^+/CCR5^+$ T lymphocytes (Paiardini *et al*., 2008). In contrast, virtually nothing is known about the role of HIV infection in the function of the oral mucosal immune system.

Oronasopharyngeal mucosal epithelia play a key role in the regulation of the immune systems at not only at the local, mucosal level but also the systemic level. The oropharyngeal mucosal tissue contains a broad-spectrum cell population representing both the adaptive and innate immune systems: $CD4^+$ and $CD8^+$ T cells and their subsets; lymphoid aggregates; and antigen-presenting cells, including Langerhans cells, macrophages, B lymphocytes, eosinophils, and mast cells (Jotwani *et al*., 2001; Zhao *et al*., 2002). Langerhans cells and macrophages in the oral epithelium have an antigen presentation function for T cells that activates an adaptive immune response (Ozbilgin *et al*., 2004). Thus, Langerhans/dendritic cells and macrophages serve as bridges between the innate and adaptive immune systems (Foti *et al*., 2004; Hoebe *et al*., 2004). The oropharyngeal epithelium may play a critical role in this bridge by regulating and enhancing the interaction of intraepithelial dendritic cells and macrophages with T-cell subsets. Oral epithelial cells may have a stronger costimulatory effect on allogeneic Langerhans cells and T-cell interactions than that of skin epithelial cells (Hasseus *et al*., 2004). Oral epithelial cells may express a number of cell surface proteins and soluble factors that may enhance the maturation of Langerhans cells and the proliferation of T cells (Hasseus *et al*., 2004). Furthermore, oral epithelial cells express

Toll-like receptors (Beklen *et al*., 2008), which might be critical for the innate immune response of oral mucosa against HIV and opportunistic viruses.

HIV may infect the immune cells in oral mucosa (Chou *et al*., 2000; Jayakumar *et al*., 2005); however, the role of HIV in dysfunction and/or modulation of the oral mucosal immune system is poorly understood. It is possible that HIV infection of oral mucosal immune cells impairs their adaptive and innate functions at the local level and therefore increases the susceptibility of oral mucosa to other opportunistic infections. For example, HIV-associated severe depletion of Langerhans cells and macrophages in the mucosal epithelium of oral hairy leukoplakia (Daniels *et al*., 1987; Tugizov *et al*., 2007) may be a critical factor permitting intensive infection by EBV in the hairy leukoplakia epithelium (Greenspan *et al*., 1985).

SUMMARY AND RECOMMENDATIONS

HIV-associated activation of opportunistic viruses in the oral cavity might be due to direct and/or indirect interactions between HIV and opportunistic viruses. Such interactions may result in a synergistic effect on the development of malignant and non-malignant oral lesions. Viral interactions within the oral mucosa may modulate cell signaling pathways that enhance the mutual activation of viruses and the reduction of the barrier and innate immune functions of the oral epithelium. Future investigation into the molecular mechanisms of viral interactions within the oral mucosa may help to reveal the pathogenic mechanisms of oral diseases and may open new possibilities for treatment of HIV-associated oral diseases. The HAART-associated infection and activation of HPV in the oral epithelium is increasing, and its molecular mechanism is not well understood. It is possible that components of HAART directly or indirectly facilitate HPV infection or activation of the oral epithelium. Investigation into the role of HAART in HPV infection, activation, and dissemination may elucidate the mechanism of HAART-associated HPV infection and HPV-related oral lesions. Oral mucosal epithelia contain epithelial and immune compartments, which play a key role in the normal functioning of the innate and adaptive immune systems at local and systemic levels. Infection by HIV and opportunistic viruses in the oral mucosa may cause complex changes in the epithelial and immune cells of oral mucosa, leading to the impairment of the oral immune system and its barrier functions. Detailed investigation of the role of HIV and opportunistic viruses in immune and barrier functions of oral mucosa may elucidate the molecular mechanism of HIV-associated susceptibility of oral mucosa to infection by opportunistic viruses.

REFERENCES

Beklen A, Hukkanen M, Richardson R, Konttinen YT (2008). Immunohistochemical localization of Toll-like receptors 1-10 in periodontitis. *Oral Microbiol Immunol* 23:425-431.

Berk T, Gordon SJ, Choi HY, Cooper HS (1985). Cytomegalovirus infection of the colon: a possible role in exacerbations of inflammatory bowel disease. *Am J Gastroenterol* 80:355-360.

Brengel-Pesce K, Morand P, Schmuck A, Bourgeat MJ, Buisson M, Bargues G, *et al*. (2002). Routine use of real-time quantitative PCR for laboratory diagnosis of Epstein-Barr virus infections. *J Med Virol* 66:360-369.

Buonaguro FM, Tornesello ML, Buonaguro L, Del Gaudio E, Beth-Giraldo E, Giraldo G (1994). Role of HIV as cofactor in HPV oncogenesis: *in vitro* evidences of virus interactions. *Antibiot Chemother* 46:102-109.

Cameron JE, Hagensee ME (2008). Oral HPV complications in HIV-infected patients. *Curr HIV/AIDS Rep* 5:126-131.

Carbone A, Cesarman E, Spina M, Gloghini A, Schulz TF (2009). HIV-associated lymphomas and gamma-herpesviruses. *Blood* 113:1213-1224.

Chaiyachati K, Cinti SK, Kauffman CA, Riddell J (2008). HIV-infected patients with anal carcinoma who subsequently developed oral squamous cell carcinoma: report of 2 cases. *J Int Assoc Physicians AIDS Care* 7:306-310.

Chang HC, Samaniego F, Nair BC, Buonaguro L, Ensoli B (1997). HIV-1 Tat protein exits from cells via a leaderless secretory pathway and binds to extracellular matrix-associated heparan sulfate proteoglycans through its basic region. *AIDS* 11:1421-1431.

Chou LL, Epstein J, Cassol SA, West DM, He W, Firth JD (2000). Oral mucosal Langerhans' cells as target, effector and vector in HIV infection. *J Oral Pathol Med* 29:394-402.

Corey L (2007). Synergistic copathogens: HIV-1 and HSV-2. *N Engl J Med* 356:854-856.

Corey L, Langenberg AG, Ashley E, Sekulovich RE, Izu AE, Douglas JM, *et al.* (1999). Recombinant glycoprotein vaccine for the prevention of genital HSV-2 infection: two randomized controlled trials. Chiron HSV Vaccine Study Group. *JAMA* 282:331-340.

Corey L, Wald A, Celum CL, Quinn TC (2004). The effects of herpes simplex virus-2 on HIV-1 acquisition and transmission: a review of two overlapping epidemics. *J Acquir Immune Defic Syndr* 35:435-445.

Daniels TE, Greenspan D, Greenspan JS, Lennette E, Schiodt M, Petersen V, *et al*. (1987). Absence of Langerhans cells in oral hairy leukoplakia, an AIDS-associated lesion. *J Invest Dermatol* 89:178-182.

Doehle BP, Hladik F, McNevin JP, McElrath MJ, Gale M Jr (2009). Human immunodeficiency virus type 1 mediates global disruption of innate antiviral signaling and immune defenses within infected cells. *J Virol* 83:10395-10405.

Ensoli B, Buonaguro L, Barillari G, Fiorelli V, Gendelman R, Morgan RA, *et al*. (1993). Release, uptake, and effects of extracellular human immunodeficiency virus type 1 Tat protein on cell growth and viral transactivation. *J Virol* 67:277-287.

Epstein JB (2007). Oral malignancies associated with HIV. *J Can Dent Assoc* 73:953-956.

Feller L, Khammissa RA, Wood NH, Meyerov R, Lemmer J (2008). Insights into immunopathogenic mechanisms of HIV infection: high levels of immune activation and HIV fitness. *SADJ* 63:552, 554, 556-557.

Fidouh-Houhou N, Duval X, Bissuel F, Bourbonneux V, Flandre P, Ecobichon JL, *et al*. (2001). Salivary cytomegalovirus (CMV) shedding, glycoprotein B genotype distribution, and CMV disease in human immunodeficiency virus-seropositive patients. *Clin Infect Dis* 33:1406-1411.

Foti M, Granucci F, Ricciardi-Castagnoli P (2004). A central role for tissue-resident dendritic cells in innate responses. *Trends Immunol* 25:650-654.

Fox-Canale AM, Hope TJ, Martinson J, Lurain JR, Rademaker AW, Bremer JW, *et al*. (2007). Human cytomegalovirus and human immunodeficiency virus type-1 co-infection in human cervical tissue. *Virology* 369:55-68.

Ganju RK, Munshi N, Nair BC, Liu ZY, Gill P, Groopman JE (1998). Human immunodeficiency virus tat modulates the Flk-1/KDR receptor, mitogen-activated protein kinases, and components of focal adhesion in Kaposi's sarcoma cells. *J Virol* 72:6131-6137.

Ghazal P, Young J, Giulietti E, DeMattei C, Garcia J, Gaynor R, *et al*. (1991). A discrete cis element in the human immunodeficiency virus long terminal repeat mediates synergistic trans activation by cytomegalovirus immediate-early proteins. *J Virol* 65:6735-6742.

Greenspan JS, Greenspan D, Lennette ET, Abrams DI, Conant MA, Petersen V, *et al*. (1985). Replication of Epstein-Barr virus within the epithelial cells of oral "hairy" leukoplakia, an AIDS-associated lesion. *N Engl J Med* 313:1564-1571.

Griffin E, Krantz E, Selke S, Huang ML, Wald A (2008). Oral mucosal reactivation rates of herpesviruses among HIV-1 seropositive persons. *J Med Virol* 80:1153-1159.

Guo X, Hutcheon AE, Zieske JD (2004). Transduction of functionally active TAT fusion proteins into cornea. *Exp Eye Res* 78:997-1005.

Hagensee ME, Cameron JE, Leigh JE, Clark RA (2004). Human papillomavirus infection and disease in HIV-infected individuals. *Am J Med Sci* 328:57-63.

Hasseus B, Jontell M, Bergenholtz G, Dahlgren UI (2004). Langerhans cells from human oral epithelium are more effective at stimulating allogeneic T cells *in vitro* than Langerhans cells from skin. *Clin Exp Immunol* 136:483-489.

Heng MC, Heng SY, Allen SG (1994). Co-infection and synergy of human immunodeficiency virus-1 and herpes simplex virus-1. *Lancet* 343: 255-258.

Herve F, Ghinea N, Scherrmann JM (2008). CNS delivery via adsorptive transcytosis. *AAPS J* 10:455-472.

Hodgson TA, Greenspan D, Greenspan JS (2006). Oral lesions of HIV disease and HAART in industrialized countries. *Adv Dent Res* 19:57-62.

Hoebe K, Janssen E, Beutler B (2004). The interface between innate and adaptive immunity. *Nat Immunol* 5:971-974.

Hyun TS, Subramanian C, Cotter MA 2nd, Thomas RA, Robertson ES (2001). Latency-associated nuclear antigen encoded by Kaposi's sarcoma-associated herpesvirus interacts with Tat and activates the long terminal repeat of human immunodeficiency virus type 1 in human cells. *J Virol* 75:8761-8771.

Jayakumar P, Berger I, Autschbach F, Weinstein M, Funke B, Verdin E, *et al*. (2005). Tissue-resident macrophages are productively infected *ex vivo* by primary X4 isolates of human immunodeficiency virus type 1. *J Virol* 79:5220-5226.

Jotwani R, Palucka AK, Al-Quotub M, Nouri-Shirazi M, Kim J, Bell D, *et al*. (2001). Mature dendritic cells infiltrate the T cell-rich region of oral mucosa in chronic periodontitis: *in situ*, *in vivo*, and *in vitro* studies. *J Immunol* 167:4693-4700.

Kakizawa J, Ushijima H, Oka S, Ikeda Y, Schroder HC, Muller WE (1996). Detection of human immunodeficiency virus-1 DNA, RNA and antibody, and occult blood in inactivated saliva: availability of the filter paper disk method. *Acta Paediatr Jpn* 38:218-223.

Kim RH, Yochim JM, Kang MK, Shin KH, Christensen R, Park NH (2008). HIV-1 Tat enhances replicative potential of human oral keratinocytes harboring HPV-16 genome. *Int J Oncol* 33:777-782.

Kim SH, Oh JM, No JH, Bang YJ, Juhnn YS, Song YS (2009). Involvement of NF-kappaB and AP-1 in COX-2 upregulation by human papillomavirus 16 E5 oncoprotein. *Carcinogenesis* 30:753-757.

Koelle DM, Posavad CM, Barnum GR, Johnson ML, Frank JM, Corey L (1998). Clearance of HSV-2 from recurrent genital lesions correlates with infiltration of HSV-specific cytotoxic T lymphocytes. *J Clin Invest* 101:1500-1508.

Lachmann R (2003). Herpes simplex virus latency. *Expert Rev Mol Med* 5:1-14.

Leigh JE, Shetty K, Fidel PL Jr (2004). Oral opportunistic infections in HIV-positive individuals: review and role of mucosal immunity. *AIDS Patient Care STDS* 18:443-456.

Levy JA (2009). HIV pathogenesis: 25 years of progress and persistent challenges. *AIDS* 23:147-160.

Lilly EA, Cameron JE, Shetty KV, Leigh JE, Hager S, McNulty KM, *et al*. (2005). Lack of evidence for local immune activity in oral hairy leukoplakia and oral wart lesions. *Oral Microbiol Immunol* 20:154-162.

Lindgren ME, Hallbrink MM, Elmquist AM, Langel U (2004). Passage of cell-penetrating peptides across a human epithelial cell layer *in vitro*. *Biochem J* 377(pt 1):69-76.

Maheshwari A, Smythies LE, Wu X, Novak L, Clements R, Eckhoff D, *et al*. (2006). Cytomegalovirus blocks intestinal stroma-induced downregulation of macrophage HIV-1 infection. *J Leukoc Biol* 80:1111-1117.

Mani D, Neil N, Israel R, Aboulafia DM (2009). A retrospective analysis of AIDS-associated Kaposi's sarcoma in patients with undetectable HIV viral loads and CD4 counts greater than 300 cells/mm(3). *J Int Assoc Physicians AIDS Care* 8:279-285.

Margolis DM, Rabson AB, Straus SE, Ostrove JM (1992). Transactivation of the HIV-1 LTR by HSV-1 immediate-early genes. *Virology* 186:788-791.

Maticic M, Poljak M, Kramar B, Tomazic J, Vidmar L, Zakotnik B, *et al*. (2000). Proviral HIV-1 DNA in gingival crevicular fluid of HIV-1-infected patients in various stages of HIV disease. *J Dent Res* 79:1496-1501.

Mbopi-Kéou FX, Bélec L, Teo CG, Scully C, Porter SR (2002). Synergism between HIV and other viruses in the mouth. *Lancet Infect Dis* 2:416-424.

McCullough MJ, Savage NW (2005). Oral viral infections and the therapeutic use of antiviral agents in dentistry. *Aust Dent J* 50(4)(suppl 2):31S-35S.

Meys R, Gotch FM, Bunker CB (2010). Human papillomavirus in the era of highly active antiretroviral therapy for human immunodeficiency virus: an immune reconstitution-associated disease? *Br J Dermatol* 162:6-11.

Milani D, Mazzoni M, Zauli G, Mischiati C, Gibellini D, Giacca M, *et al*. (1998). HIV-1 Tat induces tyrosine phosphorylation of p125FAK and its association with phosphoinositide 3-kinase in PC12 cells. *AIDS* 12:1275-1284.

Miller CS, Berger JR, Mootoor Y, Avdiushko SA, Zhu H, Kryscio RJ (2006). High prevalence of multiple human herpesviruses in saliva from human immuno-deficiency virus-infected persons in the era of highly active antiretroviral therapy. *J Clin Microbiol* 44:2409-2415.

Mosca JD, Bednarik DP, Raj NB, Rosen CA, Sodroski JG, Haseltine WA, *et al*. (1987). Activation of human immunodeficiency virus by herpesvirus infection: identification of a region within the long terminal repeat that responds to a trans-acting factor encoded by herpes simplex virus 1. *Proc Natl Acad Sci U S A* 84:7408-7412.

Mott KR, Osorio Y, Maguen E, Nesburn AB, Wittek AE, Cai S, *et al*. (2007). Role of anti-glycoproteins D (anti-gD) and K (anti-gK) IgGs in pathology of herpes stromal keratitis in humans. *Invest Ophthalmol Vis Sci* 48:2185-2193.

Nagahara H, Vocero-Akbani AM, Snyder EL, Ho A, Latham DG, Lissy NA, *et al*. (1998). Transduction of full-length TAT fusion proteins into mammalian cells: TAT-p27Kip1 induces cell migration. *Nat Med* 4:1449-1452.

Ozbilgin MK, Polat S, Mete UO, Tap O, Kaya M (2004). Antigen-presenting cells in the hypertrophic pharyngeal tonsils: a histochemical, immunuhistochemical and ultrastructural study. *J Investig Allergol Clin Immunol* 14:320-328.

Page-Shafer K, Sweet S, Kassaye S, Ssali C (2006). (C2) Saliva, breast milk, and mucosal fluids in HIV transmission. *Adv Dent Res* 19:152-157.

Paiardini M, Frank I, Pandrea I, Apetrei C, Silvestri G (2008). Mucosal immune dysfunction in AIDS pathogenesis. *AIDS Rev* 10:36-46.

Palefsky J (2009). Human papillomavirus-related disease in people with HIV. *Curr Opin HIV AIDS* 4:52-56.

Qureshi MN, Barr CE, Hewlitt I, Boorstein R, Kong F, Bagasra O, *et al*. (1997). Detection of HIV in oral mucosal cells. *Oral Dis* 3(suppl 1):73-78.

Reading PC, Whitney PG, Barr DP, Smyth MJ, Brooks AG (2006). NK cells contribute to the early clearance of HSV-1 from the lung but cannot control replication in the central nervous system following intranasal infection. *Eur J Immunol* 36:897-905.

Rebbapragada A, Wachihi C, Pettengell C, Sunderji S, Huibner S, Jaoko W, *et al*. (2007). Negative mucosal synergy between Herpes simplex type 2 and HIV in the female genital tract. *AIDS* 21:589-598.

Remeijer L, Maertzdorf J, Buitenwerf J, Osterhaus AD, Verjans GM, (2002). Corneal herpes simplex virus type 1 superinfection in patients with recrudescent herpetic keratitis. *Invest Ophthalmol Vis Sci* 43:358-363.

Rhodus NL, Ho V, Miller CS, Myers S, Ondrey F (2005). NF-kappaB dependent cytokine levels in saliva of patients with oral preneoplastic lesions and oral squamous cell carcinoma. *Cancer Detect Prev* 29:42-45.

Rodriguez-Inigo E, Jimenez E, Bartolome J, Ortiz-Movilla N, Bartolome Villar B, Jose Arrieta J, *et al*. (2005). Detection of human immunodeficiency virus type 1 RNA by *in situ* hybridization in oral mucosa epithelial cells from anti-HIV-1 positive patients. *J Med Virol* 77:17-22.

Roizman B, Whitley RJ, Lopez C, eds. (1993). *The Human Herpesviruses*. New York: Raven Press.

Ruutu M, Peitsaro P, Johansson B, Syrjanen S (2002). Transcriptional profiling of a human papillomavirus 33-positive squamous epithelial cell line which acquired a selective growth advantage after viral integration. *Int J Cancer* 100:318-326.

Sankaran S, George MD, Reay E, Guadalupe M, Flamm J, Prindiville T, *et al*. (2008). Rapid onset of intestinal epithelial barrier dysfunction in primary human immunodeficiency virus infection is driven by an imbalance between immune response and mucosal repair and regeneration. *J Virol* 82:538-545.

Santoro MG, Rossi A, Amici C (2003). NF-kappaB and virus infection: who controls whom. *EMBO J* 22:2552-2560.

Schmitz H, Rokos K, Florian P, Gitter AH, Fromm M, Scholz P, *et al*. (2002). Supernatants of HIV-infected immune cells affect the barrier function of human HT-29/B6 intestinal epithelial cells. *AIDS* 16:983-991.

Stockmann M, Schmitz H, Fromm M, Schmidt W, Pauli G, Scholz P, *et al*. (2000). Mechanisms of epithelial barrier impairment in HIV infection. *Ann N Y Acad Sci* 915:293-303.

Schwarze SR, Dowdy SF (2000). *In vivo* protein transduction: intracellular delivery of biologically active proteins, compounds and DNA. *Trends Pharmacol Sci* 21:45-48.

Tudor-Williams G, Lyall EG (1999). Mother to infant transmission of HIV. *Curr Opin Infect Dis* 12:21-26.

Tugizov S, Herrera R, Veluppillai P, Greenspan J, Greenspan D, Palefsky JM (2007). Epstein-Barr virus (EBV)–infected monocytes facilitate dissemination of EBV within the oral mucosal epithelium. *J Virol* 81:5484-5496.

Urbinati C, Mitola S, Tanghetti E, Kumar C, Waltenberger J, Ribatti D, *et al*. (2005). Integrin alphavbeta3 as a target for blocking HIV-1 Tat-induced endothelial cell activation *in vitro* and angiogenesis *in vivo*. *Arterioscler Thromb Vasc Biol* 25:2315-2320.

Vernon SD, Hart CE, Reeves WC, Icenogle JP (1993). The HIV-1 tat protein enhances E2-dependent human papillomavirus 16 transcription. *Virus Res* 27:133-145.

Vives E, Brodin P, Lebleu B (1997). A truncated HIV-1 Tat protein basic domain rapidly translocates through the plasma membrane and accumulates in the cell nucleus. *J Biol Chem* 272:16010-16017.

Yurochko AD, Kowalik TF, Huong SM, Huang ES (1995). Human cytomegalovirus upregulates NF-kappa B activity by transactivating the NF-kappa B p105/p50 and p65 promoters. *J Virol* 69:5391-5400.

Zhang RD, Guan M, Park Y, Tawadros R, Yang JY, Gold B, *et al*. (1997). Synergy between human immunodeficiency virus type 1 and Epstein-Barr virus in T lymphoblastoid cell lines. *AIDS Res Hum Retroviruses* 13:161-171.

Zhao ZZ, Savage NW, Sugerman PB, Walsh LJ (2002). Mast cell/T cell interactions in oral lichen planus. *J Oral Pathol Med* 31:189-195.