



Assessment of Oral Shedding of Herpes Simplex Virus Before and After Endodontic and Fixed Partial Prosthodontics Treatment in Patients with Recurrent Herpes Simplex

Maryam Koopaie ^{1,*}, Faezeh Esmaily² and Mohammad Javad Kharazifard^{2,1}

¹School of Dentistry, Tehran University of Medical Sciences, Tehran, Iran

²Department of Epidemiology and Biostatistics, Faculty of Public Health, Tehran University of Medical Sciences, Tehran, Iran

*Corresponding author: Assistant professor, School of Dentistry, Tehran University of Medical Sciences, Tehran, Iran. Tel: +98-9120330580, Email: mariakoopaie@gmail.com

Received 2018 July 25; Revised 2018 September 02; Accepted 2018 September 03.

Abstract

Background: Herpes simplex virus type 1 (HSV-1) as a member of the herpes virus family recurs in 25% of infected patients 1-4 times a year. Reactivation of the virus in the oral mucosa is triggered by stress, sunray, menstruation and trauma to the face.

Objectives: The current study aimed to evaluate the effect of fixed prosthetic treatment and root canal therapy on symptomatic and asymptomatic shedding of HSV-1 virus into the saliva.

Methods: Twenty two patients with a history of recurrent herpes simplex infection were selected, 11 of whom underwent endodontic treatment and 11 received fixed prosthesis treatment. Saliva samples were obtained immediately before and three days after spitting method of collection. The samples were stored at -70°C and then examined via the polymerase chain reaction (PCR) technique in order to detect probable shedding of the virus.

Results: In general, 72.7% (P=0.09) and 63.6% (P=0.09) of the samples were positive for HSV-1 in the endodontic and fixed prosthesis groups, respectively.

Conclusions: The results of this study indicated that dental treatment could be considered as a risk factor for asymptomatic recurrence of HSV-1 virus in the oral cavity. Therefore, to prevent HSV infection, the use of infection control measures and prophylactic antiviral therapy is critical in immunocompromised patients.

Keywords: PCR Test, Dental Treatment, Oral Shedding, Herpes Simplex Virus

1. Background

Herpes simplex virus type 1 (HSV-1) is a member of alpha herpes virus family that is highly infectious (1). HSV-1 is transmitted via oral-oral contact (2) and respiratory secretions (3). It is estimated that the global prevalence of HSV-1 infection in all age group is 67% (3.7 billion people), in eastern meditation countries, this prevalence is 75% (4). Most cases of HSV-1 infection are acquired during childhood as an oral infection (5).

In clinical HSV-1 infection, prodromal signs last 1-3 days and include fever, reduced appetite, weakness, diarrhea, and in some cases, headache and nausea (6, 7). General oral pain due to multiple ulcers on keratinized and non-keratinized mucosa results in reduced food intake. This disease is self-limiting and recovery takes 10 to 14 days (8, 9).

After infection, the virus starts its movement through the axon of sensory nerves, which finally causes hidden in-

fection in sensory ganglions (8). Some factors such as ultraviolet radiation, shock, stress, fever, trauma and menstruation trigger the reactivation of HSV in sensory ganglions (10, 11). As a result, the virus moves toward mucus membranes or the skin, a place where the virus can act as a pathogen for epithelial cells (12) and results in recurrent infection in the form of vesicles and topical ulcers (13) or asymptomatic shedding of virus into saliva and oral secretions (14).

Asymptomatic release of the virus is not associated with systemic signs and symptoms. There is controversy over the prevalence of asymptomatic shedding occurring after dental treatments (15-17). In some cases, HSV activation after dental procedures is very severe and can extend to the periphery of the usual site of recurrence (18, 19).

Asymptomatic shedding promotes the risk of cross-infection not only to other patients but also to dental staff (20, 21). This is especially important in immunocompro-

mised patients and pregnant women (22, 23). Therefore, the evaluation of asymptomatic HSV-1 shedding induced by dental treatment is important. In this study, we sought to determine HSV-1 oral shedding before and after dental treatments (fixed prosthesis and root canal treatment). In addition, we sought to investigate the probable effect of dental treatment on HSV-1 shedding or recurrence of HSV-1 infection.

2. Methods

Prior to the study, we obtained the approval of the Ethics Committee of Tehran University of Medical Sciences. The patients were informed of the study objectives and signed the informed consent forms.

2.1. Patients

The participants included the patients presenting to the Faculty of Dentistry of Tehran University of Medical Sciences for conventional dental care. All the patients had a history of recurrent herpes labialis (RHL). The patients were selected randomly from adults, regardless of their gender. All the patients were from the same geographical region and had comparable socioeconomic status. The exclusion criteria included immunocompromised patients, pregnant women, women in their menstrual cycle, patients presenting RHL during or seven days prior to sample collection, patients with a history of head and neck trauma, patients experiencing mental stress or acute exposure to sunlight and patients used antiviral agents during and one month prior to the study. From the 22 selected patients (m = 6, f = 16, mean age 34.57 years), 11 subjects underwent root canal therapy and the remaining 11 patients received fixed dental prosthesis treatment (Table 1).

Table 1. Demographic Data of Patient Participant to Study

| Characteristic | Endodontic | Fixed Prosthesis |
|-------------------|------------|------------------|
| Age, years | | |
| Median | 35.6 | 33.54 |
| Range | 20 - 50 | 19 - 50 |
| Gender | | |
| Female, No. (%) | 9 (81.8) | 7 (63.6) |
| Male, No. (%) | 2 (18.1) | 4 (36.3) |

2.2. Saliva Samples Collection

First, saliva samples were taken from the patients just before starting the treatment. The patients were asked to keep their saliva in their mouth for five minutes and then,

spit each 60 second until 3 ml of saliva was collected in a sterile container (spitting method). The second round of sample collection was three days after treatment using the same protocol. DNA was isolated from saliva samples using Promega DNA Purification Kit (Promega Corporation, Madison, WI, USA) according to the manufacturer's protocol and subjected to polymerase chain reaction (PCR). The samples were processed in three different physical environments at each step of the procedure to prevent contamination, namely (1) nucleic acid extraction area, (2) pre-PCR staging area, and (3) DNA amplification area, with distribution of the PCR product and detection of DNA fragments after PCR. At least three negative controls (Milli-Q water) were used per assay, including interleaving of the samples. The samples obtained from DNA extraction were submitted to amplification reaction of human β -globin DNA for the evaluation of their viability. A set of primers was designed against the HSV-1 UL42 gene (Table 2).

Table 2. Primers and PCR Reaction^a

| Virus | Primers | PCR Program |
|-------|-----------------------------|---------------------------------|
| HSV-1 | F: 5'-GCCAGCGAGACGCTGAT-3' | 94°C/60 s; 40°C/60 s; 72°C/60 s |
| | R: 5'-ACGCAGGTACTCTGGTGA-3' | |

^a F, forward primer; R, reverse primer.

2.3. Polymerase Chain Reaction (PCR)

PCR was performed using 10 μ L of 10 x PCR buffer, 4 μ L of MgC (25 mM), 8 μ L of dNTPs (2.5 mM), 2 μ L of primer HS13 (50 Pmol/ μ L), 2 μ L of primer HS14 (50 Pmol/ μ L), 1 μ L of Taq DNA polymerase (5 U/ μ L), Taq μ L of DNA sample and 63 μ L of H₂O. The UL42 gene was amplified with a 1-min denaturation step at 94°C, a 1-min annealing step at 40°C and 1 minute elongation at 72°C for 35 cycles. The PCR products were run on 2% agarose gel, stained with ethidium bromide and visualized under ultraviolet (UV) light. Positive controls of a herpesvirus culture were also included, and the negative control was water (Figure 1).

3. Results

In this cross-sectional study, 22 patients were enrolled, of whom 16 subjects were female and 6 were male. The patients' age ranged from 19 to 50 years old, and none of them had history of dental procedure-related RHL (DIRR). Eight of the 11 patients (72%; P = 0.09) undergone root canal therapy and seven of 11 patients (63%; P = 0.09) received fixed partial prosthodontics treatment had asymptomatic shedding of HSV-1. Five (45.45%) subjects from the root canal therapy group and 3 (27.3%) subjects from the

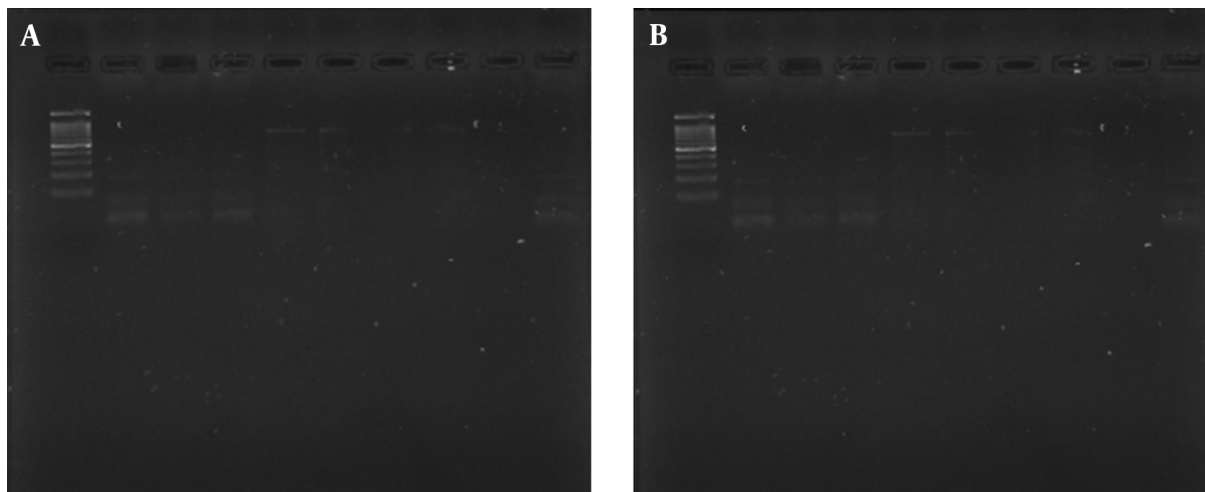


Figure 1. PCR Results: 2% agarose gel electrophoresis stained with ethidium bromide. Lane 9: negative control, lane 10: positive control. A, Endodontic treatment patient's samples; B, fixed prosthodontic treatment patient's samples.

fixed partial prosthodontics treatment group showed clinical signs of RHL or intraoral recurrent herpes infection within seven days of treatment (Figure 2). The frequency of viral shedding in patients under endodontic treatment was higher than that in the fixed prosthodontic treatment group, which could probably be due to pain and central nervous system excitement accompanied with endodontic treatment.

4. Discussion

Preliminary infection with HSV-1 can cause several health problems (24, 25). It seems that symptoms of HSV-1 shedding present three days after infection (19, 26). Thus, we collected patient specimens three days following dental treatment. PCR is an accurate molecular diagnostic tool that is very sensitive for the identification of HSV infections (27-29).

Immunoglobulins present in saliva are one of the limitations in the identification of HSV infections by cell culture, while they cannot influence PCR results (21, 30). HSV infection can be detected in about 25% of patients using viral culture and cytological methods (31). On the other hand, HSV-1 infection can be detected in 90% of patients using PCR (32, 33).

There are several studies on recurrent intraoral or labial herpes following dental treatment (20, 33). These types of studies are important because HSV shedding can cause disease transmission to dental staff and HSV keratitis and other complications of HSV infections (34, 35). In addition, HSV shedding in immunocompromised patients

may induce disseminated and potentially fatal diseases. Thus, regarding increasing HS viral load after dental treatment, considering anti-viral medications is recommended (32, 36). Miller et al. stated that at least 70% of the population shed HSV-1 asymptotically at least once a month, and many individuals appear to shed HSV-1 more than six times a month. Also, traumatic oral procedures increase the likelihood of HSV shedding in the oral cavity (33).

Hyland concluded that shedding of HSV-1 in the oral cavity not only increases by direct surgical trauma but also appears to be common in migraine and temporomandibular diseases (TMD) patients attending for general dental treatment. Thus, pain or pain-induced stress, as well as anxiety associated with dental treatment may also be a risk factor for asymptomatic shedding in specific seropositive patients receiving dental treatment (20). The study by Ramchandani et al. (14) indicated that oral reactivation of HSV-1 in comparison with HSV-1 shedding in tears and nasal mucosa is more common and usually asymptomatic. Frequent oral shedding of HSV-1 may increase the risk for transmitting the virus to oral mucosa (14); these findings confirm our results.

Asymptomatic oral HSV shedding was reported by El Hayderi et al., which was line with our findings. However, severe RHL after dental treatment was not reported in that study, whereas in our study 45.45% of root canal therapy patients and 27.3% of fixed prosthodontics patients showed RHL after dental treatment. El Hayderi et al. suggested that nerve damage due to dental treatment causes HSV-1 reactivation and concluded that oral HSV shedding may be related to invasiveness of dental procedure, but fur-

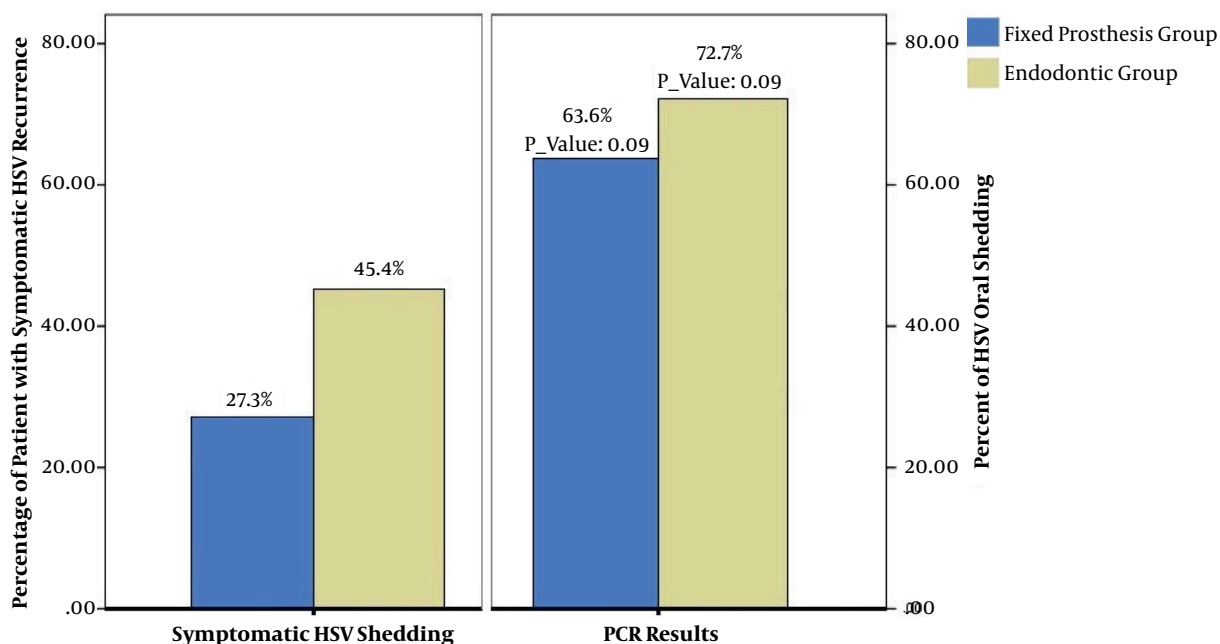


Figure 2. PCR results and percentage of symptomatic HSV shedding

ther studies are needed on this issue (2, 37).

De Santana Sarmiento et al. and Caliento et al. concluded that kidney transplant recipients (adults and children, respectively) excreted herpes viruses more often than controls, especially HSV-1 and Epstein-Barr virus, with salivary shedding of herpes viruses being more frequent in patients with recent kidney transplantation (38, 39). As we have argued elsewhere, these studies confirmed higher HSV-1 shedding in immunocompromised patients in comparison with healthy controls. The frequency of viral shedding in patients under endodontic treatment was higher than those received fixed prosthodontic treatment, which is probably due to pain and central nervous system excitement accompanied with endodontic treatment. This finding indicates that pain and pain-induced stress can activate herpes virus.

4.1. Conclusions

In this study, recurrent HSV-1 infection in 72% of the root canal and 63% of the fixed dental prosthesis patients, either symptomatic or asymptomatic, was present, which is consistent with previous reports (32, 40, 41). Asymptomatic viral shedding can be observed not only in acute infections, but also in the prodromal stage, even before clinical manifestations (2, 33). High levels of stress and dental treatment-related traumas can also elevate the risk of asymptomatic shedding in saliva (20). High levels of corti-

sol released during pain is one of the most important activators of the stress system that can instigate recurrent viral infection (42). Therefore, immunomodulation molecules that are activated due to dental treatment-related pain can cause recurrent HSV-1 infections. In addition, specific molecular signals such as adrenalin, IL-6, cAMP, glucocorticoids and prostaglandins are released during stress (42, 43).

Based on our results, the risk of viral infection transmission to dentistry staff and other patients should be strongly considered. Therefore, prophylactic acyclovir treatment before treating symptomatic patients with a history of DIRR is crucial. Overall, our results demonstrated the strong impact of dental treatment such as endodontics and prosthodontics treatment on increased HSV viral shedding in the oral cavity. An important issue is that PCR is a qualitative laboratory method; thus, determination of viral load using real time PCR is recommended.

Footnote

Ethical Considerations: IR.TUMS.VCR.REC.1395.1292.

References

1. Carratala J, Montejo M, Perez-Romero P. Infections caused by herpes viruses other than cytomegalovirus in solid organ transplant

- recipients. *Enferm Infect Microbiol Clin*. 2012;**30 Suppl 2**:63-9. doi: [10.1016/S0213-005X\(12\)70084-8](https://doi.org/10.1016/S0213-005X(12)70084-8). [PubMed: [22542037](https://pubmed.ncbi.nlm.nih.gov/22542037/)].
2. El Hayderi L, Delvenne P, Rompen E, Senterre JM, Nikkels AF. Herpes simplex virus reactivation and dental procedures. *Clin Oral Investig*. 2013;**17**(8):1961-4. doi: [10.1007/s00784-013-0986-3](https://doi.org/10.1007/s00784-013-0986-3). [PubMed: [23604785](https://pubmed.ncbi.nlm.nih.gov/23604785/)].
 3. Smith CA, Conroy LT, Pollock M, Ruddy J, Binning A, McCrudden EA. Detection of herpes viruses in respiratory secretions of patients undergoing artificial ventilation. *J Med Virol*. 2010;**82**(8):1406-9. doi: [10.1002/jmv.21794](https://doi.org/10.1002/jmv.21794). [PubMed: [20572072](https://pubmed.ncbi.nlm.nih.gov/20572072/)].
 4. Looker KJ, Magaret AS, May MT, Turner KM, Vickerman P, Gottlieb SL, et al. Global and regional estimates of prevalent and incident herpes simplex virus type 1 infections in 2012. *PLoS One*. 2015;**10**(10):e0140765. doi: [10.1371/journal.pone.0140765](https://doi.org/10.1371/journal.pone.0140765). [PubMed: [26510007](https://pubmed.ncbi.nlm.nih.gov/26510007/)]. [PubMed Central: [PMC4624804](https://pubmed.ncbi.nlm.nih.gov/PMC4624804/)].
 5. Gottlieb SL, Giersing BK, Hickling J, Jones R, Deal C, Kaslow DC, et al. Meeting report: Initial World Health Organization consultation on herpes simplex virus (HSV) vaccine preferred product characteristics, March 2017. *Vaccine*. 2017. doi: [10.1016/j.vaccine.2017.10.084](https://doi.org/10.1016/j.vaccine.2017.10.084). [PubMed: [29224963](https://pubmed.ncbi.nlm.nih.gov/29224963/)].
 6. Stoopler ET, Balasubramaniam R. Topical and systemic therapies for oral and perioral herpes simplex virus infections. *J Calif Dent Assoc*. 2013;**41**(4):259-62. [PubMed: [23705241](https://pubmed.ncbi.nlm.nih.gov/23705241/)].
 7. Tovar S, Parlatescu I, Tovar M, Cionca L, Arduino PG. Recurrent intraoral HSV-1 infection: A retrospective study of 58 immunocompetent patients from Eastern Europe. *Med Oral Patol Oral Cir Bucal*. 2011;**16**(2):e163-9. [PubMed: [20711124](https://pubmed.ncbi.nlm.nih.gov/20711124/)].
 8. Burket LW, Greenberg MS, Glick M. *Burket's oral medicine: Diagnosis and treatment*. PMPH-USA; 2003.
 9. Regezi JA, Sciubba JJ, Jordan RCK. *Oral pathology: Clinical pathologic correlations*. illustrated ed. Elsevier; 2016.
 10. Kennedy PG, Rovnak J, Badani H, Cohrs RJ. A comparison of herpes simplex virus type 1 and varicella-zoster virus latency and reactivation. *J Gen Virol*. 2015;**96**(Pt 7):1581-602. doi: [10.1099/vir.0.000128](https://doi.org/10.1099/vir.0.000128). [PubMed: [25794504](https://pubmed.ncbi.nlm.nih.gov/25794504/)]. [PubMed Central: [PMC4635449](https://pubmed.ncbi.nlm.nih.gov/PMC4635449/)].
 11. Whitley RJ. Herpes simplex virus infections of the central nervous system. In: Peterson P, Toborek M, editors. *Neuroinflammation and Neurodegeneration*. New York, NY: Springer; 2014. p. 263-90.
 12. Agelidis AM, Shukla D. Cell entry mechanisms of HSV: what we have learned in recent years. *Future Virol*. 2015;**10**(10):1145-54. doi: [10.2217/fvl.15.85](https://doi.org/10.2217/fvl.15.85). [PubMed: [27066105](https://pubmed.ncbi.nlm.nih.gov/27066105/)]. [PubMed Central: [PMC4822157](https://pubmed.ncbi.nlm.nih.gov/PMC4822157/)].
 13. Horowitz R, Aierstuck S, Williams EA, Melby B. Herpes simplex virus infection in a university health population: clinical manifestations, epidemiology, and implications. *J Am Coll Health*. 2010;**59**(2):69-74. doi: [10.1080/07448481.2010.483711](https://doi.org/10.1080/07448481.2010.483711). [PubMed: [20864431](https://pubmed.ncbi.nlm.nih.gov/20864431/)].
 14. Ramchandani M, Kong M, Tronstein E, Selke S, Mikhaylova A, Magaret A, et al. Herpes simplex virus type 1 shedding in tears and nasal and oral mucosa of healthy adults. *Sex Transm Dis*. 2016;**43**(12):756-60. doi: [10.1097/OLQ.0000000000000522](https://doi.org/10.1097/OLQ.0000000000000522). [PubMed: [27835628](https://pubmed.ncbi.nlm.nih.gov/27835628/)]. [PubMed Central: [PMCS117635](https://pubmed.ncbi.nlm.nih.gov/PMCS117635/)].
 15. Arduino PG, Porter SR. Herpes simplex virus type 1 infection: overview on relevant clinico-pathological features. *J Oral Pathol Med*. 2008;**37**(2):107-21. doi: [10.1111/j.1600-0714.2007.00586.x](https://doi.org/10.1111/j.1600-0714.2007.00586.x). [PubMed: [18197856](https://pubmed.ncbi.nlm.nih.gov/18197856/)].
 16. McGeoch DJ, Bowden RJ. Evolution of herpes simplex viruses. *Herpes simplex viruses*. CRC Press; 2017. p. 1-34.
 17. Szpara ML, Gatherer D, Ochoa A, Greenbaum B, Dolan A, Bowden RJ, et al. Evolution and diversity in human herpes simplex virus genomes. *J Virol*. 2014;**88**(2):1209-27. doi: [10.1128/JVI.01987-13](https://doi.org/10.1128/JVI.01987-13). [PubMed: [24227835](https://pubmed.ncbi.nlm.nih.gov/24227835/)]. [PubMed Central: [PMC3911644](https://pubmed.ncbi.nlm.nih.gov/PMC3911644/)].
 18. Simon E, Matee M. Post-extraction complications seen at a referral dental clinic in Dar Es Salaam, Tanzania. *Int Dent J*. 2001;**51**(4):273-6. [PubMed: [11570541](https://pubmed.ncbi.nlm.nih.gov/11570541/)].
 19. Arduino PG, Porter SR. Oral and perioral herpes simplex virus type 1 (HSV-1) infection: review of its management. *Oral Dis*. 2006;**12**(3):254-70. doi: [10.1111/j.1601-0825.2006.01202.x](https://doi.org/10.1111/j.1601-0825.2006.01202.x). [PubMed: [16700734](https://pubmed.ncbi.nlm.nih.gov/16700734/)].
 20. Hyland PL, Coulter WA, Abu-Ruman I, Fulton CR, O'Neill HJ, Coyle PV, et al. Asymptomatic shedding of HSV-1 in patients undergoing oral surgical procedures and attending for noninvasive treatment. *Oral Dis*. 2007;**13**(4):414-8. doi: [10.1111/j.1601-0825.2007.01316.x](https://doi.org/10.1111/j.1601-0825.2007.01316.x). [PubMed: [17577329](https://pubmed.ncbi.nlm.nih.gov/17577329/)].
 21. Fatahzaadeh M, Schwartz RA. Human herpes simplex virus infections: epidemiology, pathogenesis, symptomatology, diagnosis, and management. *J Am Acad Dermatol*. 2007;**57**(5):737-63. quiz 764-6. doi: [10.1016/j.jaad.2007.06.027](https://doi.org/10.1016/j.jaad.2007.06.027). [PubMed: [17939933](https://pubmed.ncbi.nlm.nih.gov/17939933/)].
 22. Tsai J, Cohrs RJ, Nagel MA, Mahalingam R, Schmid DS, Choe A, et al. Reactivation of type 1 herpes simplex virus and varicella zoster virus in an immunosuppressed patient with acute peripheral facial weakness. *J Neurol Sci*. 2012;**313**(1-2):193-5. doi: [10.1016/j.jns.2011.08.040](https://doi.org/10.1016/j.jns.2011.08.040). [PubMed: [21924743](https://pubmed.ncbi.nlm.nih.gov/21924743/)]. [PubMed Central: [PMC3243805](https://pubmed.ncbi.nlm.nih.gov/PMC3243805/)].
 23. Kucera P, Gerber S, Marques-Vidal P, Meylan PR. Seroepidemiology of herpes simplex virus type 1 and 2 in pregnant women in Switzerland: an obstetric clinic based study. *Eur J Obstet Gynecol Reprod Biol*. 2012;**160**(1):13-7. doi: [10.1016/j.ejogrb.2011.09.030](https://doi.org/10.1016/j.ejogrb.2011.09.030). [PubMed: [21982938](https://pubmed.ncbi.nlm.nih.gov/21982938/)].
 24. Wu YP, Sun DD, Wang Y, Liu W, Yang J. Herpes simplex virus type 1 and type 2 infection increases atherosclerosis risk: Evidence based on a meta-analysis. *Biomed Res Int*. 2016;**2016**:2630865. doi: [10.1155/2016/2630865](https://doi.org/10.1155/2016/2630865). [PubMed: [27195284](https://pubmed.ncbi.nlm.nih.gov/27195284/)]. [PubMed Central: [PMC4853930](https://pubmed.ncbi.nlm.nih.gov/PMC4853930/)].
 25. Campadelli-Fiume G, De Giovanni C, Gatta V, Nanni P, Lollini PL, Menotti L. Rethinking herpes simplex virus: the way to oncolytic agents. *Rev Med Virol*. 2011;**21**(4):213-26. doi: [10.1002/rmv.691](https://doi.org/10.1002/rmv.691). [PubMed: [21626603](https://pubmed.ncbi.nlm.nih.gov/21626603/)].
 26. Westley S, Seymour R, Staines K. Recurrent intra-oral herpes simplex 1 infection. *Dent Update*. 2011;**38**(6):368-70. 372-4. doi: [10.12968/denu.2011.38.6.368](https://doi.org/10.12968/denu.2011.38.6.368). [PubMed: [21905349](https://pubmed.ncbi.nlm.nih.gov/21905349/)].
 27. Tateishi K, Toh Y, Minagawa H, Tashiro H. Detection of herpes simplex virus (HSV) in the saliva from 1,000 oral surgery outpatients by the polymerase chain reaction (PCR) and virus isolation. *J Oral Pathol Med*. 1994;**23**(2):80-4. [PubMed: [8164158](https://pubmed.ncbi.nlm.nih.gov/8164158/)].
 28. Sacks SL. Famciclovir suppression of asymptomatic and symptomatic recurrent anogenital herpes simplex virus shedding in women: a randomized, double-blind, double-dummy, placebo-controlled, parallel-group, single-center trial. *J Infect Dis*. 2004;**189**(8):1341-7. doi: [10.1086/383038](https://doi.org/10.1086/383038). [PubMed: [15073669](https://pubmed.ncbi.nlm.nih.gov/15073669/)].
 29. da Silva LM, Guimaraes AL, Victoria JM, Gomes CC, Gomez RS. Herpes simplex virus type 1 shedding in the oral cavity of seropositive patients. *Oral Dis*. 2005;**11**(1):13-6. doi: [10.1111/j.1601-0825.2004.01058.x](https://doi.org/10.1111/j.1601-0825.2004.01058.x). [PubMed: [15641961](https://pubmed.ncbi.nlm.nih.gov/15641961/)].
 30. Kaufman HE, Azcuy AM, Varnell ED, Sloop GD, Thompson HW, Hill JM. HSV-1 DNA in tears and saliva of normal adults. *Invest Ophthalmol Vis Sci*. 2005;**46**(1):241-7. doi: [10.1167/iovs.04-0614](https://doi.org/10.1167/iovs.04-0614). [PubMed: [15623779](https://pubmed.ncbi.nlm.nih.gov/15623779/)]. [PubMed Central: [PMC1200985](https://pubmed.ncbi.nlm.nih.gov/PMC1200985/)].
 31. Gitman MR, Ferguson D, Landry ML. Comparison of Simplexa HSV 1 & 2 PCR with culture, immunofluorescence, and laboratory-developed TaqMan PCR for detection of herpes simplex virus in swab specimens. *J Clin Microbiol*. 2013;**51**(11):3765-9. doi: [10.1128/JCM.01413-13](https://doi.org/10.1128/JCM.01413-13). [PubMed: [24006008](https://pubmed.ncbi.nlm.nih.gov/24006008/)]. [PubMed Central: [PMC3889765](https://pubmed.ncbi.nlm.nih.gov/PMC3889765/)].
 32. Miller CS, Avdiushko SA, Kryscio RJ, Danaher RJ, Jacob RJ. Effect of prophylactic valacyclovir on the presence of human herpesvirus DNA in saliva of healthy individuals after dental treatment. *J Clin Microbiol*. 2005;**43**(5):2173-80. doi: [10.1128/JCM.43.5.2173-2180.2005](https://doi.org/10.1128/JCM.43.5.2173-2180.2005). [PubMed: [15872238](https://pubmed.ncbi.nlm.nih.gov/15872238/)]. [PubMed Central: [PMC1153765](https://pubmed.ncbi.nlm.nih.gov/PMC1153765/)].
 33. Miller CS, Danaher RJ. Asymptomatic shedding of herpes simplex virus (HSV) in the oral cavity. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 2008;**105**(1):43-50. doi: [10.1016/j.tripleo.2007.06.011](https://doi.org/10.1016/j.tripleo.2007.06.011). [PubMed: [17703961](https://pubmed.ncbi.nlm.nih.gov/17703961/)].
 34. Browning WD, McCarthy JP. A case series: herpes simplex virus as an occupational hazard. *J Esthet Restor Dent*. 2012;**24**(1):61-6. doi: [10.1111/j.1708-8240.2011.00469.x](https://doi.org/10.1111/j.1708-8240.2011.00469.x). [PubMed: [22296698](https://pubmed.ncbi.nlm.nih.gov/22296698/)]. [PubMed Cen-

- tral: [PMC3437498](#)].
35. Tsatsos M, MacGregor C, Athanasiadis I, Moschos MM, Hossain P, Anderson D. Herpes simplex virus keratitis: an update of the pathogenesis and current treatment with oral and topical antiviral agents. *Clin Exp Ophthalmol*. 2016;**44**(9):824-37. doi: [10.1111/ceo.12785](#). [PubMed: [27273328](#)].
 36. Klein RS. *Prevention of herpes simplex virus type 1 infection in immunocompetent patients*. Up To Date, Inc; 2016.
 37. El Hayderi L, Raty L, Failla V, Caucanas M, Paurobally D, Nikkels AF. Severe herpes simplex virus type-1 infections after dental procedures. *Med Oral Patol Oral Cir Bucal*. 2011;**16**(1):e15-8. [PubMed: [20526251](#)].
 38. De Santana Sarmiento DJ, Tozetto-Mendoza TR, Masami Sumita L, Pierroti LC, Pallos D, Caliento R, et al. Oral shedding of human herpesviruses in renal transplant recipients. *Clin Oral Investig*. 2018;**22**(2):885-91. doi: [10.1007/s00784-017-2166-3](#). [PubMed: [28669106](#)].
 39. Caliento R, Sarmiento DJS, Silva EMP, Tozetto-Mendoza TR, Toubouti PL, Benini V, et al. Oral shedding of HSV-1 and EBV and oral manifestations in paediatric chronic kidney disease patients and renal transplant recipients. *Acta Odontol Scand*. 2018:1-6. doi: [10.1080/00016357.2018.1437218](#). [PubMed: [29409366](#)].
 40. Marques-Silva L, Castro WH, Gomez EL, Guimaraes AL, Silva MS, Gomez RS. The impact of dental surgery on HSV-1 reactivation in the oral mucosa of seropositive patients. *J Oral Maxillofac Surg*. 2007;**65**(11):2269-72. doi: [10.1016/j.joms.2007.05.029](#). [PubMed: [17954324](#)].
 41. Gaudin RA, Remenschneider AK, Phillips K, Knipfer C, Smeets R, Heiland M, et al. Facial palsy after dental procedures - Is viral reactivation responsible? *J Craniomaxillofac Surg*. 2017;**45**(1):71-5. doi: [10.1016/j.jcms.2016.11.002](#). [PubMed: [27939042](#)].
 42. Sainz B, Loutsch JM, Marquart ME, Hill JM. Stress-associated immunomodulation and herpes simplex virus infections. *Med Hypotheses*. 2001;**56**(3):348-56. doi: [10.1054/mehy.2000.1219](#). [PubMed: [11359358](#)].
 43. Ives AM, Bertke AS. Stress Hormones Epinephrine and Corticosterone Selectively Modulate Herpes Simplex Virus 1 (HSV-1) and HSV-2 Productive Infections in Adult Sympathetic, but Not Sensory, Neurons. *J Virol*. 2017;**91**(13). doi: [10.1128/JVI.00582-17](#). [PubMed: [28404850](#)]. [PubMed Central: [PMC5469259](#)].