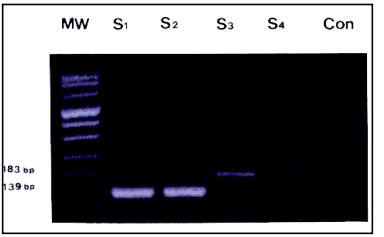

Figure 1 - Detection of amplification products using 4% gel and ethidium bromide staining. The 139 bp fragment corresponds to *Leishmania* amplified DNA and the 183 bp fragment corresponds to internal control amplified DNA. MW - molecular weight standers, S1, S2, S4 - positive samples of visceral leishmaniasis, S3 - negative sample Con - internal control.

(**Figure 1**), that all (4/4) were negative by microscopy and cultivation, probably due to the low number of *Leishmania* parasites in the aspirates.

Despite the recent advancement and development of medical services in Syria within the last few years, VL is still endemic and considered one of the challenges confronting the medical profession. The presence of VL parasites in dogs as well as in *Phlebotomus tobbi* (vector), in areas around the humid and sub-humid coastal zone and the surrounding mountains in Syria was demonstrated.[5] Indeed, Idlep and Lattakia Governorates are located within this zone, and exhibited the highest prevalence. Dry arid regions such as Damascus and inland provinces maintained the lowest number of reported cases. Due to the high sensitivity and specificity of the PCR technique,[6,7] it was used to diagnose suspected patients who presented the clinical symptoms of VL but its parasite could not be demonstrated by biological tests,[2,3] which shows the parasites microscopically, in stained smears, *in vitro* cultivation and indirectly by serological means,[3] whereas the PCR test depends upon the detection of *Leishmania* DNA in VL suspected samples. The kDNA was composed of up to 10000 copies of approximately 800 bp minicircles.[8] The multicopy target sequence combined with a simplified sample preparation procedure, allowed the detection of low levels of *Leishmania* parasites, with the results obtained within 24 hours.[1] The PCR of bone marrow aspirates results were positive while the microscopic examination and *in vitro* cultivation results were both negative, which confirm the high sensitivity and specificity of the PCR technique.

In conclusion, the PCR technique is more sensitive than the traditional diagnostic tests and especially useful for confirmation of VL, as an endemic disease in Syria, which should receive more attention from the health authorities and the health professionals in the country.

Acknowledgment. We are very grateful to Dr. Y. Le Fichoux (Chu, France) for his advice and for enabling meaningful testing of the PCR methodology. We further thank our clinical colleagues who collected patient samples and clinical evaluation.

Received 25th August 2004. Accepted for publication in final form 12th December 2004.

From the Department of Animal Biology, Faculty of Sciences, Damascus University, Damascus, Syria. Address correspondence and reprint request: Dr. Samar Al-Nahhas, Professor, Faculty of Sciences, Damascus University, PO Box 10718, Damascus, Syria. Tel. +963 (11) 3716872. Fax. +963 (11) 3732338. E-mail: samar@scs-net.org

References

1. Brecelj M, Pikelg F, Gubensek f, Anderluh G. Polymerase chain reaction as a diagnostic tool for detecting *Leishmania*. ***Infection*** 2000; 28: 111-113.
2. Mathis A, Deplazes P. PCR and in vitro cultivation for detection of *Leishmania spp.* In diagnostic samples from humans and dogs. ***J Clin Microbiol*** 1995; 33: 1145-1149.
3. Schalling HDFH, Schoone GJ, Kroon CCM, Hailu A, Chappuis F, Vecken H. Development and application of simple diagnostic tools for VL. ***Med Microbiol Immunol*** 2001; 190: 69-71.
4. Riera C, Fisa R, Udina M, Galego M, Portus M. Detection of *Leishmania infantum*, cryptic infection in asymptomatic blood donors living in an endemic area (Evissa; Balearic Islands; Spain) by different diagnostic, methods. ***Trans R Soc Trop Med Hyg*** 2004; 98: 102-110.
5. Rioux JA, Leger N, Haddad N, Gramiccia M, Jalouk L, Dereure T, et al. Infestation naturelle de *Phlebotomus tobbi (Diptera - Psychodidae) par Leishmania donovani .s. st. (Kintoplastidae – Trypanosomatidae)* en Syrie. ***Parassitologia*** 1998; 40: 148.
6. Singh S, Sivakumar R. Recent advances in the diagnosis of leishmaniasis. ***J Postgrad Med*** 2003; 49: 55-60.
7. Osman OF, Oskam L, Kroon NCM, Schoone GJ, Khalil ETAG, El-Hassan AM, et al. Use of PCR for diagnosis of Post-Kala-azar dermal Leishmaniasis. ***J Clin Microbiol*** 1998; 36: 1621-1624.
8. Lambson B, Smyth A, Barker DC. *Leishmania donovani* development and characterization of a kinetoplaste DNA Probe and its use in the detection of parasites. ***Exp Parasitol*** 2000; 94: 15-22.

Vitiligo and human herpesvirus 6. *Is there a relationship?*

***Ali K. Adiloglu**, MD, **Pinar Y. Basak**, MD, **Rabia Can**, MD, **Vahide B. Akkaya**, MD.*

Vitiligo is the acquired loss of melanocytes leading to areas of depigmentation. It affects approximately 1% of the population. Among

vitiligo etiology theories, autoimmune theory remains the most popular one. Latent viral infections have been postulated to be the triggering factors in the development of autoimmune diseases. Human herpesvirus 6 (HHV-6) attacks the cell nucleus, it is double-stranded and often remains dormant for many years and are large enough to scatter ultraviolet light. Human herpesvirus 6 demonstrates predominantly CD4+ cell trophism. Similar to HHV-6, at the onset of vitiligo the main lymphocytes are CD4 T-lymphocytes and vitiligo causes alterations in T-lymphocyte subsets, an aberrant natural killer cell activity and antibody dependent cell-mediated cytotoxicity.[1] We aimed to study the role of HHV-6 in the etiology of vitiligo. To our knowledge there is no study in determining the relation between vitiligo and HHV-6 so far.

Eighty vitiligo patients with 80 age and gender matched controls who have minor dermatological problems were included in the study from those admitted to the Dermatology Department of the University Hospital. Patients' age, gender, duration of illness, age at onset of the disease, course of vitiligo as active or stable, type of vitiligo and percentage of involvement and treatment received were recorded. All subjects provided signed informed consent after approval of the experimental protocol by the Medical School Ethics Committee. Serum anti-HHV-6 immunoglobulin M (IgM) and immunoglobulin G (IgG) antibodies were measured by enzyme-linked immunosorbent assay (ELISA) method (Panbio Inc., Maryland, United Stated of America) and quantitative serum C-reactive protein (CRP) levels were measured by Behring nephelometry system using particle enhanced immunonephelometry method (N High Sensitivity CRP, Behring, Germany). Statistical analyses were performed using Statistical Package for Social Sciences for Windows (version 10). Categorical data of sera results were analyzed by chi-square test. Fisher's exact test was used for smaller groups. Independent samples T-test were used for comparison of quantitative sera results.

Of the 80 patients recruited 37 (46.3%) were male and 43 (53.7%) were female. The mean age of patients was 39.6 ± 16.7 (10 - 84 years), the mean age at onset of the disease was 30.3 ± 16.7 (4 - 78 years), the mean duration of the illness was 9.3 ± 10.6 (1 - 40 years) years. Thirty-three of patients (41.3%) had active vitiligo and 47 (58.7%) had stable vitiligo. Sixty-four patients (80%) had less than 25% involvement, 11 patients (13.7%) had between 25-50%, 4 patients (5%) had between 50-75% and one patient (1.3%) had more than 75% involvement. Thirty-seven patients (46.2%) were diagnosed as localized, 9 (11.3%) were segmental, 21 (26.3%) were acral or acrofacial and 13 (16.2%) were generalized types of vitiligo. Twenty-eight (35%) patients received systemic psoralen plus ultraviolet radiation (PUVA), 2 (2.5%) patients received local PUVA, and 18 (22.5%) patients were treated with local corticosteroid treatment. Thirty-two (40%) patients did not receive any treatment at all. Anti-HHV-6 IgG seropositivities were detected in 66 (82.5%) of 80 patients and 49 (61.3%) of 80 controls and the difference was statistically significant (p=0.003, χ^2= 8.93). Anti HHV-6 IgG positivity ratio was significantly higher in segmental vitiligo patients (13/13; 100%) compared with acral and acrofacial (12/19; 63.2%) involvement (p=0.0252, odd's ratio=16.2) (Fisher's exact test). Anti-HHV-6 IgM was positive in 9 (11.3%) of patients and 5 (6.3%) of controls, which was not enough for a significant relation. Six of 33 (18.2%) active vitiligo patients were CRP positive whereas only 4 of 47 (8.5%) of stable vitiligo patients were CRP positive but the difference was not statistically significant (p=0.198, χ^2=1.658).

Human herpesvirus 6 is the major cause of roseola infantum. Supposed roles for HHV-6 in several diseases, which are linked to viral infections have been reported; multiple sclerosis, infectious mononucleosis-like illness, lymphadenopathy, lymphoma, leukemia, pityriasis rosea, oral and cervical cancer, gloves and socks syndrome, Gianotti-Crosti syndrome and severe drug eruptions.[2] The relation between HHV-6 and vitiligo has not been studied up to now. In our study, HHV-6 IgG seropositivity was found to be higher in vitiligo group compared with the control group. All HHV-6 IgM positive patients were also HHV-6 IgG positive. The prevalence of HHV-6 IgG level in the healthy group (61.3%) was similar to previous studies. The prevalence of HHV-6 in different populations ranged from 63.5% to 88.1%.[3,4] To our knowledge, there has been no epidemiological study detecting HHV-6 seropositivity in healthy population in our country. In our study, the mean value of quantitative CRP levels of patients and controls did not differ significantly and the mean serum CRP levels did not differ between HHV-6 IgG or IgM seropositives and seronegatives in patient group. C-reactive protein is synthesized from hepatocytes after acute inflammation or tissue destruction and it is not diagnostic for specific diseases and increases with any kind of inflammation. C-reactive protein was also found to increase in vasculitis and in atherosclerotic coronary artery diseases[5] in which there is chronic infection. These studies imply that acute or chronic infections increase CRP levels. Although chronic infections have been suggested to initiate vitiligo, there may not be a substantial amount of inflammation enough to increase serum CRP levels in our study. The relation of vitiligo and microorganisms other than herpesviruses has also been studied. Grimes et al[6] studied herpes simplex virus (HSV), varicella zoster, cytomegalovirus

(CMV), Epstein-Barr virus, HIV and human T-cell lymphotrophic virus DNA in skin biopsy specimens in 29 vitiligo patients and 22 control subjects and only CMV-DNA was identified in 38% of the patients, whereas all control subjects were negative. Intercellular adhesion molecule-1 (ICAM-1) on the surface of epidermal keratinocytes and melanocytes is likely to greatly influence cytotoxic damage of these cells in diseases like photosensitive lupus erythematosus, lichen planus, erythema multiforme, and vitiligo. It is proposed that disease-specific induction of ICAM-1 by factors such as ultraviolet radiation and herpesvirus infection, is an important determinant in triggering these skin diseases and in determining the pattern of disease.[7] Our results also comply with the immunologic proposal of vitiligo as herpesviruses may trigger ICAM-1 expression on melanocytes which may activate autoimmune destruction of these cells.

In conclusion, HHV-6 IgG seropositivity showing past HHV-6 infection is related to vitiligo. C-reactive protein, which is elevated in acute or chronic inflammation and tissue damage, is related neither to vitiligo nor to HHV-6. Human herpesvirus 6 infection in a genetically susceptible host could potentially mediate the destruction of melanocytes by induction of aberrant humoral and cell-mediated immunological responses eventually causing vitiligo. Nevertheless, advanced immunologic and pathologic proofs of viral infection in skin is needed to confirm vitiligo-virus relationship.

Received 21st October 2004. Accepted for publication in final form 12th December 2004.

From the Departmentof Microbiology and Clinical Microbiology (Adiloglu, Can) and the Department of Dermatology (Basak, Akkaya), Suleyman Demirel University Medical School, Isparta, Turkey. Address correspondence and reprint requests to Dr. Ali K. Adiloglu, Posta Kutusu 61, 32100, Isparta, Turkey. Tel. +90 (246) 2324811. Fax. +90 (312) 4250760. E-mail: aadiloglu@yahoo.com

References

1. De Araujo T, Berman B, Weinstein E. Human herpesviruses 6 and 7. ***Dermatol Clin*** 2002; 20: 301-306.
2. Kosuge H. Human herpesvirus-6, 7 and their related diseases. ***J Dermatol Science*** 2000; 22: 205-212.
3. Carricart SE, Bustos DA, Grutadauria SL, Nates SV, Garcia JJ, Yacci MR, et al. Human herpesvirus 6 circulation in healthy adults and oncologic patients. ***Medicina*** 2002; 62: 9-12.
4. Bhattarakosol P, Pancharoen C, Mekmullica J, Bhattarakosol P. Seroprevalence of anti-human herpes virus-6 IgG antibody in children of Bangkok, Thailand. ***Southeast Asian J Trop Med Public Health*** 2001; 32: 143-147.
5. Ridker PM, Hennekens CH, Buring IE, Rifai N. C- reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. ***N Engl J Med*** 2000; 342: 836-843.
6. Grimes PE, Ghoneum M, Stockton T, Payne C, Kelly AP, Alfred L. T cell profiles in vitiligo. ***J Am Acad Dermatol*** 1986; 14: 196-201.
7. Norris DA. Cytokine modulation of adhesion molecules in the regulation of immunologic cytotoxicity of epidermal targets. ***J Invest Dermatol*** 1990; 95 (6 Suppl): 111-120.

Use of laryngeal mask airway for the care of rhinoplasty

Jamil S. Anwari*, MBBS, FFARCS,* ***Javaid Bhatti****, MBBS.*

Some operations on the face, such as rhinoplasty, require care to preserve the delicate surgical work. In anesthetized patient after extubation, firm application of face mask (such as Rush™) is required for adequate spontaneous or manual ventilation. Pressure exerted on the nose by the face mask may change the shape of newly reconstructed nose. Subcutaneous emphysema has been reported after rhinoplasty where air was pumped through the lateral osteotomy incision.[1,2] The risk of emphysema would probably increase if patient receives positive pressure ventilation via face mask. Awake extubation is often associated with straining or bucking increasing the nasal bleeding, which is undesirable in this situation. To avoid both tracheal extubation response and face mask ventilation, the trachea can be extubated during deep anesthesia and ventilation can be maintained with laryngeal mask airway (LMA). As compared to Guedal airway, LMA provides easier airway maintenance.[3] Use of LMA after extubation during emergence from anesthesia compared with awake extubation or extubation in anesthetized without LMA, has been associated with less respiratory complications during recovery period.[4] Laryngeal tube has been used during emergence from anesthesia in a patient with an unstable neck.[5]

We present results on 15 cases of rhinoplasties where at the end of the operation patients were extubated during deep anesthesia and ventilation was maintained through LMA until patients regained consciousness. Mean age of patients were 32 years [standard deviation (SD) of 5] and mean body weight is 68 kg (SD = 8.7), and they were either American Society of Anesthesiologist class I or II. All our patients received oral pre-medication of diazepam and metoclopramide 10 mg each orally 2 hours before the operation. After establishing IV line and standard monitoring, general anesthesia was induced with fentanyl (2 μg/kg), thiopentone (3-5 mg/kg) and cisatracurium (0.5 mg/kg). Patients were ventilated with face mask until all 4 twitches of train of 4 disappeared, and were then intubated. Anesthesia was maintained with sevoflurane, nitrous oxide and oxygen. Additional narcotic (morphine 1.5 mg/kg, intramuscularly), nonnarcotic