Oral Focal Epithelial Hyperplasia: Report of Five Cases

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Focal epithelial hyperplasia or Heck’s disease is a rare contagious disease caused by human papillomavirus types 13 or 32, initially described among Native American populations. This condition is characterized by the occurrence of multiple small papules or nodules in oral cavity, especially on labial and buccal mucosa and tongue. This report describes the diagnosis of focal epithelial hyperplasia in five Central Amazonian Indians who sought treatment at the Amazonas State Foundation of Tropical Medicine (FMT-AM), using clinical criteria, polymerase chain reaction (PCR) and DNA sequencing.

Key Words: focal epithelial hyperplasia, polymerase chain reaction, South American Indians.

INTRODUCTION

Focal epithelial hyperplasia (FEH) or Heck’s disease is a rare contagious disease caused by human papillomavirus that was first described in 1965 from the observation of isolated or multiple soft papular and nodular eruptions on the oral mucosa of Navajo Xavante Indian and Alaska Eskimo children (1). Focal epithelial hyperplasia was initially reported mostly among Native Americans, Eskimos and South Africans but has also been found in other ethnic groups. It is more frequent in younger age groups and sometimes has also a characteristic familial occurrence, which led to the suggestion that a genetic predisposition may contribute to the development of the disease (2).

FEH manifests on the mucosa as multiple or unique soft papules of whitish or normal color with a smooth surface and measuring 1 to 10 mm in diameter. The lesions are painless, tend to disappear spontaneously, and are predominantly found on the lower lip, buccal mucosa and tongue, and less often on the upper lip, gingiva and palate (2,3).

The etiologic agent of FEH was first characterized in 1983 and was designated as HPV 13, which is related to HPV 6 and HPV 11 (4). Years later, another type of HPV was isolated from FEH and referred as HPV 32, which was also related to HPV 6 and HPV 13 (5). More recently, FEH was renamed as multifocal papillomavirus epithelial hyperplasia (MPVEH) (6).

This paper reports five typical cases of focal epithelial hyperplasia and demonstrates the association with HPV 13 through polymerase chain reaction (PCR) and sequencing of PCR products.

CASE REPORT

Five patients (3 male and 2 female, aged 3 to 17 years) from a Central Amazonian Indian community sought treatment at the Amazonas State Foundation of Tropical Medicine (FMT-AM) in Manaus (Brazil). They
presented multiple soft papular whitish to normally
colored mucosal lesions, predominantly located on the
lower lip and buccal mucosa, but also affecting the
upper lip and tongue mucosa (Fig. 1). Two of the
patients were siblings. The lesions were asymptomatic
and the only discomfort they caused was their presence
itself. All lesions were classified as papular, according
to Pilgard’s criteria (7). The clinical diagnosis of FEH
was straightforward, and main differential diagnosis
included other lesions caused by HPV, especially
condylomata acuminata. To confirm the diagnosis,
superficial epithelial cells were collected from the lesions
for molecular examination. To avoid unnecessary invasive
techniques and discomfort to the patients, no histological
examination or other types of tests were performed until
the results of molecular analysis were obtained.

**PCR Analysis and DNA Sequencing**

Sample collection: cells from mucosal lesions
were collected by the cytobrush technique, immersed in
400 μL of TE buffer (Tris-HCl 10 mM pH 8.0 and EDTA
1 mM) and stored at -20°C until use (8).

Preparation of samples for PCR: samples were
digested in a solution with 2% Tween 20, 200 μg/mL of
proteinase K in TE (Tris-HCl 10mM pH 8.0 and EDTA
1 mM), incubated at 55°C for 1 h and then at 95°C for
10 min. The digested samples were maintained at -20°C
until use (8).

Amplification reaction: the universal primers
MY09 and MY11 were used to amplify L1 region of
HPV 450 bp fragments. Primer sequence was MY09 -
5'CGT CCM AAR GGA WAC TGA TC3' and MY11 -
5'GCM CAG GGW CAT AAY AAT GG3' (8).

PCR conditions: a typical reaction system
containing a final volume of 50 μL was composed of 5
μL DNA sample; 2.5 mM MgCl2, 0.25 mM dNTP, 25
pmol of each primer MY09 and MY11, 4U Taq DNA
polymerase, 50 mM KCl and 10 mM Tris-HCl (pH 8.5).
PCR reactions were carried out using an Eppendorf
Master Cycler Gradient thermocycler (Eppendorf
Scientific, Inc., Westbury, NY, USA), according to the
following protocol: 95°C for 2 min (Hot-Start),
denaturation at 95 °C for 1 min, annealing at 55°C for
1 min and polymerization at 72°C for 1 min (35 cycles),
followed by end extension at 72°C for 5 min. The
products were analyzed in 1.5% agarose gel
electrophoresis.

**DNA Sequencing**

PCR products were purified using a
MicroSpinTM S-400 column (Amersham Biosciences
Corp., Piscataway, NJ, USA). The purified PCR products
were used for sequencing following the protocol: 1 μL
of primer MY11 (upstream) 25 pmol, 4 μL of mix
Dynamic ET terminator cycle sequencing kit (Amersham
Biosciences Corp.) and 5 μL of purified PCR products.
The sequencing reaction was carried out using the
Eppendorf Master Cycler Gradient thermocycler
(Eppendorf Scientific, Inc.), according to the following
cycling profile: 25 s at 95°C (Hot-start), 15 s at 95°C for
denaturation, 10 s at 50°C for annealing and extension
at 60°C for 1 min (35 cycles). Finally, the sequencing
products were precipitated with 0.1V of ammonium
acetate 7.5 M and 4V of ethanol absolute for analysis.
The sequencing purified products were dissolved in 10
μL of Loading buffer (70% formamide and 1 mM
EDTA).

The analysis of sequencing purified products
were performed in the MegaBACE 1000 automated
sequencing system (Amersham Biosciences Corp.) and
then sequences were compared with GeneBank HPV
sequences (www.ncbi.nlm.nih.gov) using FASTA
sequences and BLAST program.

**RESULTS**

All samples were positive for HPV and showed
450 bp DNA fragments by electrophoresis on 1.5%
agarose gel, corresponding to the HPV L1 capsid protein (Fig. 2).

The analysis of sequencing purified products with BLAST program showed 99% of similarity with HPV 13 (gi | 60295 | emb | X62843.1 |) (9).

**DISCUSSION**

Focal epithelial hyperplasia does not seem to be a diagnostic challenge as long as careful examination and description of the lesions are made and patient’s medical history is comprehensively reviewed. Important data are those regarding communal way of life, with characteristic sharing of food, personal objects and lack of hygiene, which is typically observed among Brazilian Indian communities. The lesions themselves are also quite easily identifiable because of their multiplicity, small diameter of each isolated papule or nodule, soft consistence and typical intraoral topography. According to some authors, lesion predilection for lip, buccal and tongue mucosa is a sign of the infectious nature of the disease and is consistent with the patients’ communal way of life (10).

The lesions have soft consistence and discreet size, being predominantly manifested as small-diameter flat papules rather than elevated nodules, which is a consequence of their typical histopathologic architecture. All alterations occur in the epithelial layer of the mucosa, with virtually no alteration in the underlying connective tissue (11). As the molecular analysis allowed a conclusive diagnosis no histological examination was performed for diagnostic purposes in the cases presented in this report.

Setting the diagnosis of FEH is extremely important because of the need for the differential diagnosis with other conditions, namely inflammatory fibrous hyperplasia, inflammatory papillary hyperplasia, verruciform xanthoma, verrucous carcinoma, Cowden’s disease, condyloma acuminatum, and focal dermal hypoplasia syndrome (Goltz-Gorlin syndrome) (2). The first three lesion types mentioned above are reactive lesions and, in most cases, an irritating agent can be identified. Verrucous carcinoma is a neoplasm that occurs in a different age group, with epidemiological features typically found in oral carcinomas. Cowden’s disease, characteristic of an older age group, presents fibroepithelial polyps, which are more consistent, less mobile and have different intraoral topography.

A differential diagnosis with condyloma acuminatum is important because the clinical appearance of isolated lesions in both diseases is similar, as they are both caused by HPV. In spite of this, the patient’s medical history is very helpful for differentiation, and FEH lesions tend to be flatter and more numerous. In addition, the location of FEH lesions (lip, tongue and buccal mucosa) is very characteristic.

PCR is a useful tool to identify the viral etiology of FEH lesions because it is a rapid and sensitive method (8). An additional advantage of PCR using consensus primers to HPV detection is the range of viral diversity that can be identified. Once the presence of HPV was detected in the cases presented in this paper, sequencing of PCR products was important to establish which viral type was actually the etiologic agent of FEH (in these cases, HPV-13). Thus, the differential diagnosis of condyloma acuminatum is rejected, as well as other papillomatous infections caused by HPV, such as laryngeal papillomatosis (12). The molecular results are consistent with the epidemiological features of the patients, as well as with the clinical characteristics of the lesions.

Interestingly, none of the cases were positive for HPV-32 by DNA sequencing. HPV-32 is another type of HPV that causes FEH (5). No conclusions can be drawn about the prevalence of HPV-13 or -32 infection in the patients’ community because only five cases have been studied.

FEH is described in the literature as a benign
condition that heals spontaneously and therefore requires no treatment, except in some cases of functional (e.g., lesions that are constantly traumatized on biting) or aesthetic impairment (12). In the cases presented, all patients were treated. A 3-yr-old patient had habit of sucking the lesions and other patients had lesions that were constantly traumatized on biting. Several treatment modalities have been proposed for FEH, as scalpel surgery, cryotherapy, laser ablation, cauterization and topical treatments with retinoic acids or interferon (13). In the patients hereby described, topical treatments were excluded because the patients were not cooperative and compliant enough. Among the surgical treatments available in our service, cauterization was chosen because it presents lesser risks of bleeding and infection. Healing of all cases was uneventful. Follow-up of these patients is important for evaluation of treatment success. Likewise, epidemiological studies with the larger Indian groups are urged to know the exact prevalence of this disease among these populations, the long-term behavior of the lesions and the treatment protocols that might be required.

REFERENCES


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