Journal of the Formosan Medical Association (2013) 112, 756–760

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.jfma-online.com

ORIGINAL ARTICLE

Langerhans cells in odontogenic epithelia of odontogenic fibromas

Yang-Che Wu [a,b], Yi-Ping Wang [a,b], Julia Yu-Fong Chang [a,b,c], Hsin-Ming Chen [a,b], Andy Sun [a,b], Chun-Pin Chiang [a,b,d,*]

[a] *Graduate Institute of Clinical Dentistry, School of Dentistry, National Taiwan University, Taipei, Taiwan*
[b] *Department of Dentistry, National Taiwan University Hospital, College of Medicine, National Taiwan University, Taipei, Taiwan*
[c] *Department of Oral and Maxillofacial Surgery, Division of Oral Pathology, School of Dentistry, University of Washington, Seattle, USA*
[d] *Graduate Institute of Oral Biology, School of Dentistry, National Taiwan University, Taipei, Taiwan*

Received 5 September 2013; received in revised form 29 October 2013; accepted 5 November 2013

KEYWORDS
CD1a;
immunohistochemistry;
Langerhans cell;
odontogenic epithelium;
odontogenic fibroma;
S-100 protein

Background/Purpose: Langerhans cell (LC) is an antigen-presenting cell that is very important for T-cell-mediated immune reactions. Our previous studies have shown the presence of LCs in some odontogenic tumors and cysts. In this study, we further examined the presence of LCs in odontogenic epithelia of 16 odontogenic fibromas (OFs).
Methods: Anti-CD1a and anti-S-100 immunostains were used to detect the presence of LCs in nests or strands of odontogenic epithelia of 16 OFs.
Results: These 16 OFs included 10 peripheral OFs excised from seven male and three female patients (mean age, 38 years) and six central OFs (including one recurrent OF) removed from five male patients (mean age, 28 years). Of the 10 peripheral OFs, six were found on the mandibular gingiva and four on the maxillary gingiva. Four central OFs were located in the maxilla and two in the mandible. We found that both anti-CD1a and anti-S-100 immunostains had an equal ability to identify LCs in OFs. Positively stained dendritic LCs could be detected in nests and strands of odontogenic epithelia in nine (six peripheral and three central OFs) of the 16 OFs. In five peripheral OFs, dendritic LCs were found in occasional nests or strands of odontogenic epithelia. In one peripheral and three central OFs, dendritic LCs could be detected in at least half of the nests or strands of odontogenic epithelium in the tissue section.
Conclusion: LCs can be detected in the nests or strands of odontogenic epithelia in approximately 60% of the 10 peripheral OFs and approximately 50% of the six central OFs detected.

Conflicts of interest: none.

* Corresponding author. Department of Dentistry, National Taiwan University Hospital, Number 1, Chang-Te Street, Taipei 10048, Taiwan.
E-mail address: cpchiang@ntu.edu.tw (C.-P. Chiang).

0929-6646/$ - see front matter
http://dx.doi.org/10.1016/j.jfma.2013.11.002

Introduction

Odontogenic fibroma (OF) is a rare odontogenic tumor characterized by varying numbers of nests or strands of odontogenic epithelium in a mature fibrous stroma. OFs can further be divided into central (intraosseous) and peripheral (extraosseous) OFs according to the anatomic sites involved.[1] Our previous study has reported the clinicopathological features of 12 peripheral OFs and three central OFs in 15 Taiwanese patients. The peripheral OFs seem to have a predilection for the mandibular gingiva (67%).[2]

Langerhans cell (LC) is an antigen-presenting cell that is very important for T-cell-mediated immune reactions. LCs can be demonstrated by anti-S-100 or anti-CD1a immunostain in odontogenic epithelia of odontogenic tumors such as the noncalcifying variant of Pindborg tumor,[3–5] classical Pindborg tumor,[6] central granular cell odontogenic tumor,[7,8] ameloblastoma,[9,10] and OF,[11,12] as well as in the lining epithelia of radicular cysts,[10.13] dentigerous cysts,[10,13] odontogenic keratocysts,[9,13] and calcifying odontogenic cysts.[9] Because only a few OF cases have been examined for the presence of LCs in odontogenic epithelia of OFs and the results are controversial, this study further used anti-CD1a and anti-S-100 immunostains to assess the presence of LCs in the nests or strands of odontogenic epithelia of 16 OFs.

Materials and methods

The study group consisted of 16 OF cases retrieved from the archives of the Department of Oral Pathology and Oral Diagnosis, National Taiwan University Hospital, Taipei, Taiwan, from January 2006 to December 2012. Of these 16 OF cases, four (Cases 1, 2, 3, and 11 in Table 1) had been reported previously.[2] Demographic data, including gender and age of patients, as well as location, size, clinical diagnosis, treatment, and recurrence of the lesions were obtained by reviewing the dental or medical charts.

All surgical specimens were obtained from total excision or enucleation of the tumor. The specimens were then fixed in 10% neutral formalin for at least 24 hours, dehydrated in graded alcohol, and then embedded in paraffin. The tissue blocks were cut in serial sections of 4 μm thickness, and stained with hematoxylin and eosin. The histopathological diagnosis was confirmed by examination of the hematoxylin and eosin-stained tissue sections. Microscopic criteria for the diagnosis of OFs included the presence of nests or strands of odontogenic epithelium and foci of osteoid-, cementoid-, or cementicle-like calcifications in a fibrous or myxomatous stroma. Because the clinicopathological findings of 12 peripheral OFs and three central OFs have already been reported in our previous study,[2] this study focused on the evaluation of the presence of LCs in nests or strands of odontogenic epithelia in 16 OFs.

Immunohistochemistry for studying LCs

All the specimens for immunohistochemical staining were fixed in 10% neutral formalin, embedded in paraffin, and cut in serial sections of 4 μm thickness. Immunohistochemical staining was performed using a supersensitive polymer-horseradish peroxidase technique. Briefly, tissues sections were deparaffinized and rehydrated. Next, they were placed in a plastic slide holder (Dako, Copenhagen, Denmark) containing 0.01 M citrate buffer, heated in a microwave oven for 15 minutes to retrieve antigenicity, and then treated with 3% H_2O_2 in methanol for 10 minutes to quench endogenous peroxidase activity. After washing in 10 mM Tris-buffered saline (TBS), pH 7.6, the sections were incubated with 10% normal goat serum (BioGenex, San Ramon, CA, USA) to block nonspecific binding. These sections were then incubated overnight at 4°C with mouse anti-S-100 monoclonal antibody (Thermo Fisher Scientific, Runcorn, UK) at a dilution of 1:100 or mouse anti-CD1a monoclonal antibody (Abcam plc, Cambridge, UK) at a dilution of 1:50. BioGenex Super Sensitive TM detection systems were used for the detection of bound antibodies. After washing in TBS, the sections were treated with a super enhancer reagent for 10 minutes and subsequently with the polymer-horseradish peroxidase reagent for another 10 minutes. Diaminobenzidine hydrochloride (0.02%) (Zymed Laboratories, San Francisco, CA, USA) containing 0.03% H_2O_2 was used as a chromogen to visualize peroxidase activity. The preparations were counterstained lightly with hematoxylin, mounted with Permount, and examined by light microscopy. Sections of a human melanoma and a human LC histiocytosis that were previously shown to be positive for S-100 and CD1a proteins, respectively, were used as positive controls. TBS instead of a primary antibody was used as a negative control.

LCs in our samples were considered positive for S-100 or CD1a protein expressions when a prominent brown cytoplasmic staining was observed within nests or strands of odontogenic epithelium. Initially, the sections were scanned at low power. For sections that showed heterogeneous patterns of positive staining, the predominant pattern was taken into account for scoring. At least five high-power (200×) fields were chosen randomly, and the number of foci of positively stained LCs in nests or strands of odontogenic epithelium was counted for each case. The mean number of positively stained LC foci per high-power field was counted and graded as follows: −, no positively stained LC focus; +, ≤3 positively stained LC foci per high-power field; and ++, >3 positively stained LC foci per high-power field. Each of these assessments was carried out by two investigators independently. In this study, the interobserver reproducibility was 100%.

Results

Demographic and clinical data of 16 OF cases in 15 patients as well as the mean number of positively stained LC foci detected by anti-CD1a and anti-S-100 immunostains in 16 OF specimens are presented in Table 1. Case 16 was a recurrent central OF of Case 15, which recurred 8 months after the initial excision of the tumor. Other 14 OF cases showed no recurrence of the lesion after a follow-up period ranging from 10 months to 79 months. The 16 OFs included 10 peripheral OFs excised from seven male and three female patients (mean age, 38 years) and six central OFs removed from five male patients (mean age, 28 years). Of