上顎骨孤立型形質細胞瘤免疫組織化學偵測及超微構造之探討——病例報告

黃湧澧　林立民　陳中和
林正仲　印憶恆

形質細胞瘤（plasma cell neoplasm）可分為多發性骨髓瘤（multiple myeloma）、孤立型形質細胞瘤（solitary plasmacytoma）及髓質外形質細胞瘤（extramedullary plasmacytoma）。孤立型形質細胞瘤極其罕見，為形質細胞於局部骨骼之惡性增殖而未漫延至其它骨骼系統。本文報告一發生於上顎骨的形質細胞瘤，放射線檢查發現於上顎骨左側後方有一囊腫樣骨破壞性病灶。電腦斷層放射線檢查顯示此種塊已侵犯左側顏臉頰部、上顎竇及鼻腔外側壁。病理組織檢查發現此病灶為一形態變化少且緻密排列的圓形及多角形細胞組成，腫瘤細胞有巨大、偏心位置的細胞核而似形質細胞。腫瘤細胞之超微構造顯示許多呈平行排列的顆粒性內質網及豐富的高基氏體，許多沿細胞核周圍環繞的粒腺體，細胞核染色質呈濃縮斑塊及有巨大的核仁。由抗氧化酶（peroxidase antiperoxidase）免疫組織化學法分析發現病變細胞呈單株型染色（anti-Kappa 鏈抗體及 anti-Ig G），此反應顯示形質細胞為腫瘤性增殖，腫瘤經由外科手術摘除後並接受總劑量為 4200 RADS 的放射線治療，術後三年追蹤檢查顯示患者一般情況良好，無腫瘤復發現象。

Key words: plasmacytoma, ultrastructure, immunoperoxidase staining

(Kaohsiung J Med Sci 6: 565–571, 1990)

發生在顎骨及口腔軟組織的形質細胞（plasma cell）病變略可以分成四大類[1,2]：(1)多發性髓質瘤（multiple myeloma），(2)骨內孤立型形質細胞瘤（solitary plasmacytoma of bone），(3)髓質外形質細胞瘤（extramedullary plasmacytomas），(4)形質細胞肉芽腫（plasma cell granuloma）。

其中髓質瘤及形質細胞瘤為腫瘤性增殖的形質細胞組成，可發生於許多部位之骨骼或僅出現在單一部位之骨骼，但也有出現於單一骨骼病變後繼發於身體其它部位之情形，且病灶波及至顎骨常可能為多發性髓質瘤之表徵之一，曾有報告指出受侵犯部位包括上顎骨[3]、下顎骨[4,5]，齒齦[6]、唾液腺[7,8]，及頸部之淋巴節[9]。形質細胞瘤臨床上的特性與其細胞組織形態上的差異無關[10]，有些細胞瘤為良性病灶在完全摘除後即不再復發，但有些卻具有局部侵犯性，而另有些甚至造成其它骨骼的侵犯而與多發性髓質瘤較難區分。在口腔組織中通常伴隨有炎性病灶所造成的形質細胞緻密聚集的形質細胞肉芽腫與形質細胞瘤，在組織病理學上不易作鑑別診斷，腫瘤性（neoplastic）與反應性（reactive）形質細胞的增殖病灶，於正確的診斷下，其治療的成功率卻有相當大的差別[10]。

腫瘤性增殖之形質細胞的組成，其所產生之單一型球蛋白，於多發性髓質瘤時，可由患者血清偵測出來；但是孤立型形質細胞瘤及髓

高雄醫學院　牙醫學系

質外形質細胞瘤所產生的單一型球蛋白，則因合成量不足，故未能於患者血清偵測出。在反應性形質增殖病患，則有多型式的免疫球蛋白（polytypic immunoglobulins）之形成。

本文報告一例發生於上顎骨之孤立型形質細胞瘤，並藉由免疫組織化學法偵測病變細胞內免疫球蛋白（intracelluar immunoglobulins）的種類來鑑別腫瘤性或反應性形質細胞，並探討電子顯微鏡所見形態學上的表現。

病例報告

(A)病人：

病患為42歲男性於數月前因左側顏面區腫脹，至牙醫診所求診並接受骨放射線檢查，發現在其左側上顎後牙區有一放射線透過性病灶，臨床診斷疑似囊腫樣骨破壞性病灶。經由保守性外科摘除術，手術過程順利，但是由於病灶深及上顎骨深部，而無法將此囊腫樣病灶完全摘除。此腫塊於術後一個月急速腫大且疼痛而佔據整個傷口區，於是轉診至高雄醫學院附設中和紀念醫院牙科做進一步檢查。口內檢查所見為一4×3公分直徑大小、顏色為暗咖啡色的突起腫塊（Fig. 1），左側上顎前庭亦被腫塊破壞，整個腫塊所侵犯的範圍由左側第一小臼齒至第二大臼齒區，環口X光片檢查有不規則囊腫樣骨破壞性放射線透過病灶，同時在上顎粗隆有兩顆贅生齒（Fig. 2），電腦斷層放射線檢查此突起腫塊侵佔了鼻腔外側壁、顏臉頰側及上顎竇（Fig. 3）。其餘臨床常規檢查皆正常，骨掃描及身體其它部位骨骼系統X光檢查亦無疑似病變區，尿液生化檢查無Bence-Jones蛋白質沈澱，及免疫球蛋白質電泳檢查亦屬正常範圍，最後診斷為上顎骨孤立型形質細胞瘤，腫瘤經由外科手術摘除後並接總劑量為4200 RADS的放射線治療，術後三年追蹤檢查顯示一般情況良好，無腫瘤復發現。

(B)特殊檢查：

（I）免疫組織化學法：

主要以Sternberger創立之抗過氧化酶法[11]，將病灶組織標本固定於10%福馬林溶液

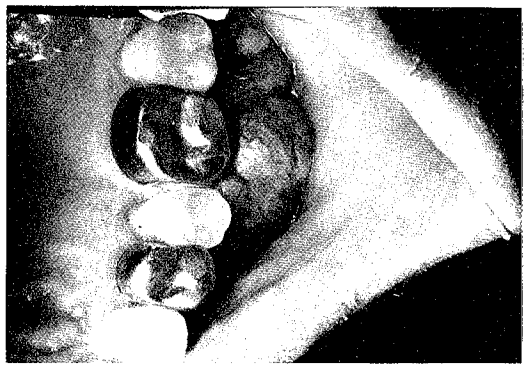


Fig. 1. Intraoral examination revealed a bulgy mass of the left maxillary area; invading from the upper second premolar to the second molar. The left buccal maxillary vestibule was obliterated by the swelling mass.

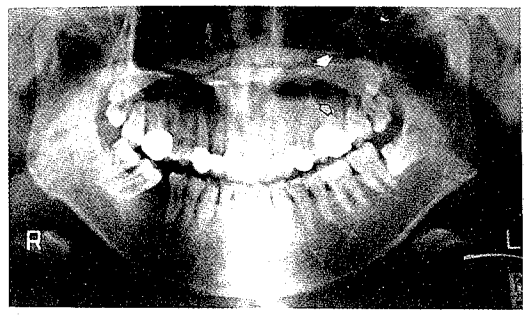


Fig. 2. Panoramic view shows a cystic-like osteolytic radiolucency with irregular defined border invading from second premolar to the left maxillary tuberosity, with two supernumerary teeth.

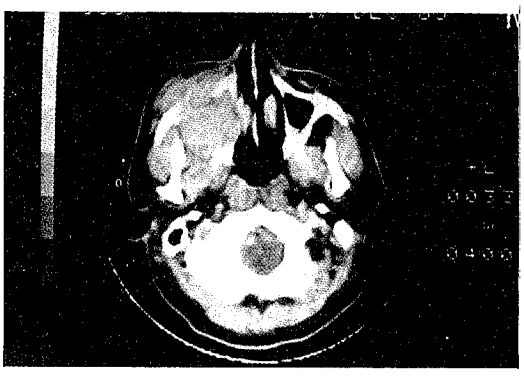


Fig. 3. CT scan, coronal section through maxillary antrum, showing the tumor mass extending toward the buccal surface of face, maxillary sinus, and lateral wall of nasal cavity.

中，臘塊包埋並切成4 μm之厚度附於塗有3%之Elmer膠水之載玻片，經過脫臘後，以3%Hydrogen peroxide-methanol（1：4）之試液滴浸於切片上且置於潮濕盒內30分鐘，以阻斷組織內所含有之內在性過氧化酶（endogenous peroxidase），之後置pH 值7.4，TRIS緩衝鹽水清洗10分鐘；再以天鵝血清（swine serum）處理30分鐘，以阻斷非特異性免疫球蛋白之吸附（non-specific immunoglobin adsorption），將過多的血清擦拭乾淨；切片再與1：50稀釋之兔子抗人體免疫球蛋白抗體作用，包括rabbit-anti-human kappa及Lambda輕鏈、Ig G、Ig A、Ig M及Ig D，於潮濕盒內30分鐘，再置於TRIS緩衝鹽水沖洗；繼而以Peroxidase-antiperoxidase複合體處理30分鐘，置於緩衝鹽水完全清洗乾淨；再以DAB（6 μg／10 ml TRIS緩衝鹽水加上100 μl 3% H_2O_2）為染色原（chromogen）處理50分鐘，置於流水中清洗10分鐘，再以蘇木精（hematoxylin）為對照染色，脫水澄清，蓋上蓋玻片，以普通光學顯微鏡觀察。

（Ⅱ）電子顯微鏡觀察：

以穿射式電子顯微鏡方法，將一部份檢體立即切取數小塊0.1至0.2公分之組織，先固定在含2%paraformaldehyde及2.5%glutaraldehyde之0.1M Cacodylate緩衝鹽水1小時，再以含0.1%Osmium tetraoxide的0.1M Cacodylate緩衝鹽水2小時，再經由酒精系列脫水，Epikoto包埋，以Porter-Blum MT2 Ultramicrotome做成超薄切片，再經由uranyl acetate及lead citrate雙重染色，用JOLE200型電子顯微鏡觀察，並攝影記錄。

結　　果

（Ⅰ）組織病理所見：

檢體組織標本體積為7.5×6.0×4.0公分，呈暗紅棕色的腫塊（Fig. 4），是取於左側上顎犬齒近心側至上顎粗隆之病灶區，檢體表面為多葉狀平滑外形，其腹面為上顎骨分隔腔，同時在後上方有兩顆贅生齒。檢體組織在光學鏡下主要是由緻密堆積之圓形或多角形細胞所組成，每一個腫瘤細胞含一巨大、單一偏心位置的細胞核，其細胞質呈半月型（cresent），核染色質在細胞核內排列呈鐘面型，似非典型之形質細胞分佈於稀疏基質中，部份的腫瘤細胞呈大小不一，分佈不均勻的不正常形質細胞，亦偶而可發現有雙葉狀細胞核之非典型形質細胞（Fig. 5）。最後病理斷為上顎骨孤立型形細胞瘤。

（Ⅱ）免疫組織化學所見：

腫瘤細胞之細胞質與antil-Lambda鍵呈陰性反應，而與anti-Kappa鍵呈陽性反應（Figs. 6a，6b），抗免疫球蛋白檢驗，則只對anti-IgG呈陽性反應。

（Ⅲ）電子顯微鏡所見：

腫瘤細胞為一橢圓形，表面光滑並且具有微絨毛，細胞胞核相當大且呈偏心位置，核仁明顯、核膜結構完整。濃密異染質呈斑塊狀分

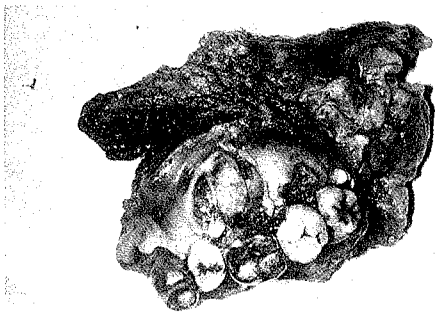


Fig. 4. The gross specimen of partial maxillectomy, the resected mass was 7.5 x 6.0 cm in size, dark brownish in color, extending from the upper first premolar to the third molar area.

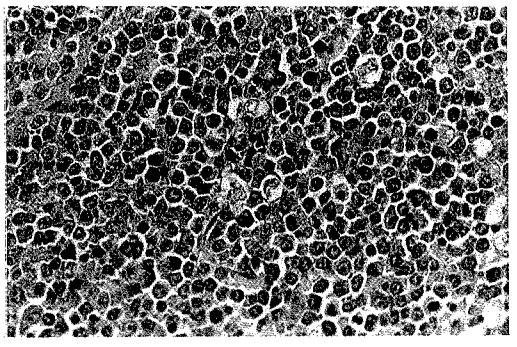
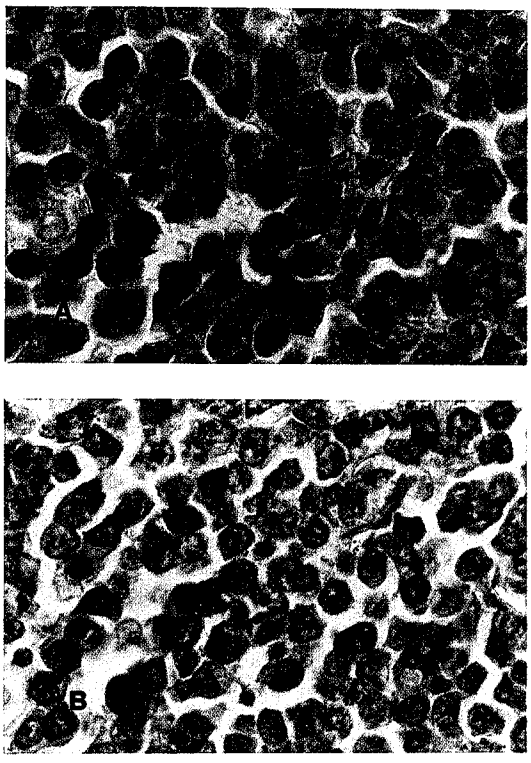


Fig. 5. Light microscopic examination reveals that the tumor cells consist of densely packed, uniform appearance round and polygonal cells. The eccentric nucleus has "clock-face" chromatin materials and sparse stroma, resembling plasma cells (H&E, X100).



Figs. 6a, 6b. PAP immunohistochemical examination reveals dark brownish staining of cytoplasm, of all plasma cells showing positively, for the human Kappa light chain and IgG respectively (a); the photomicrograph showed the negative staining of plasma cells for the human Lamda light chain, IgA, IgM, and Ig D (b) (Mayer's hematoxylin counterstain; X400).

佈於核內且與較淡的染色質分佈呈類似輪輻的形狀（Fig. 7a）。細胞質內含豐富擴張條狀的顆粒性內質網，有些呈層狀排列，但亦有些呈菊紋狀排列（Fig. 7b）。細胞核周圍區有許多圓型或長型的粒腺體，及光滑膜小泡（vacuoles）的高基氏體；在高基氏體的附近有膜被顆粒（membrane-bounded granules）（Fig. 7c）。偶而亦可以發現雙葉狀的細胞核（bilobulated nucleus）及不正常分裂現象（Fig. 7d），具有明顯層狀排列之顆粒性內質網及豐富之大型粒腺體。腫瘤性形質細胞亦存有緻密電子之包涵體（inclusion body），由全部或部份平滑膜所環繞，有的呈現律性節條狀、管狀晶體結構，亦有部份之包涵體可以於細胞外發現（Fig. 7e）。部份細胞核內有osmophilic 輕電子緻密度之無形物質（Fig. 7f）。

討　　論

孤立型形質細胞瘤之所以被認為是一種骨髓之腫瘤，是因其侵犯骨髓腔而造成瘤性腫脹之特性，且經常波及皮質骨變薄而導至病理性骨折。腫瘤細胞主要侵犯胸骨、肋骨、脊椎骨及顎骨，而長骨為較不易發生之部位[12]。

臨床上診斷為孤立型形竊細胞瘤的準據，Bataille 及 Sany[12] 提出六項要件：一、經由放射線檢查、骨掃或電腦斷層攝影證實為孤立型骨腫瘤（即無其他骨骼被侵犯），二、活體組織切片證實為形質細胞腫瘤性增殖，三、骨髓檢查中，缺乏形質細胞存在，四、無貧血、高血鈣症或腎臟病變，五、無單株抗體成分或降低抗體成分，六、正常的球蛋白量。本病例符合以上條件，確定診斷為孤立型形質細胞瘤。

形質細胞瘤為放射線敏感且癒後良好的病灶[13-14]。所以臨床上治療以放射治療為主。為防止其再復發通常採用高劑量之放射線治療[13-16]，（4000-6000 RADS），對於局部腫塊體積相當大且侵犯速度快者，對於治療癒後情況則不良，且有較高的局部復發率，長時期的追蹤是必要的。本病例接受上顎半顎切除腫瘤及總劑量為4200 RADS 的術後放射線治療且牙科補綴物亦完全適應，而且病灶並無再發的現象。經過了三年的追蹤檢查，目前生活的情形非常良好。

本病例之大部分腫瘤增殖性形質細胞具有斑塊濃縮的核染色質，巨大的核仁及相當完備的核膜，與以往學者[17-21]的發現一致。斑塊濃縮的核染色質使得細胞呈輪輻狀，此於一般正常形質細胞較不易發現，Smetana 等[21]認為分裂時期核仁的存在與形質細胞的合成核糖核酸有關，並且在細胞核周圍有豐富的粒腺體，可供給能量以利核糖核酸的輸送，因此形質細胞具有較高的代謝率（metabolic rate）或不正常的代謝作用。

正常形質細胞細胞質內有層狀排列的顆粒性內質網，而免疫球蛋則積沈於擴張性內質網槽內，本病例亦發現有許多膜被球狀顆粒於高基氏體附近，這些被認為是濃縮球蛋白或勒塞耳小體（Russell bodies）。管狀晶體結構（

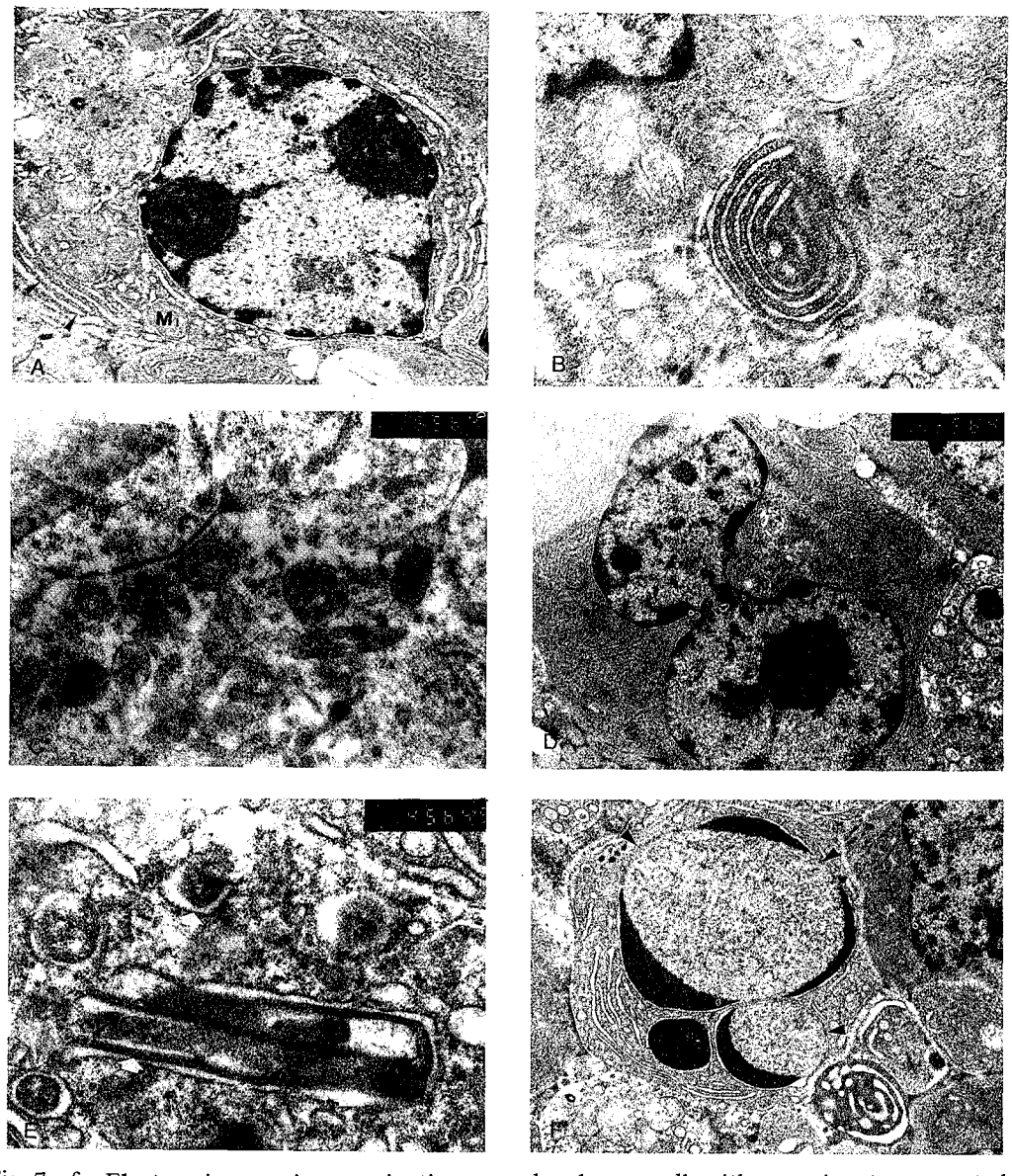


Fig. 7a-f. Electromicroscopic examination reveals plasma cell with prominent compacted nucleous and patchy condensation of nuclear chromatin. The cytoplasm contains numerous dilated strands of rough endoplamic reticulum, arranging in a lamellar fashion (arrows) and the mitchondria (Mi) distributing in the perinuclear region (a; x 13,600); Annular and semicircular rough endoplasmic reticulum in the cytoplasm of the plasma cell is found (b; x 28,900); Some spherical membrane-bounded, dense granules are presented in the Golgi complex and adjacent area (c; x 40,500); The bilobulated nucleus exhibits the "cartwheel" appearance of heterochromatin. The rough endoplasmic reticulum is well-developed and consists of flat parallel cisternae (d; x 14,600); The inclusion bodies are completely or partially encircled by a smooth membrane. A linear periodicity can be seen in some of the inclusion bodies (arrows) (e; x 31,500); The nucleus of plasma cell exhibits two single membrane-bounded, inclusion bodies, which contain slightly electron-dense amorphous osmophilic material (arrows) (f; x 10,200).

tubular crystalline structure）被認為是球蛋白的沈澱處[17-22]，此結構存在於細胞內的內質網槽內，本研究發現於細胞內、外有管狀晶體結構，與其他學者報告[22-23]之功能性髓質細胞（functional myeloma cell）具有相同形態學的表現，這可能與本病例臨床上極高度活躍性的腫瘤性增殖有密切的相關性。

腫瘤細胞由抗氧化酶免疫組織化學法對anti-Kappa及anti-IgG呈現單株性染色之陽性反應，與Taylor等[24]報告認為腫瘤性增殖為單株性生長相符合。增殖細胞源自單一種來源的起始細胞（single progenitor），更可以證實本病例的形質細胞為腫瘤性增殖，而非多株性的反應性增殖。本研究利用免疫組織化學方法，確認病灶區內形質細胞是否為單株或多株性增殖，為鑑別診斷形質細胞是否為腫瘤性增殖或反應性增殖的有效工具。

REFERENCES

1. Shafter WG, Hine MK, Levy BM: A Textbook of Oral Pathology. WB Saunders Co., Philadelphia, 3rd ed.: 178–181, 1974.
2. Bhaskar SN, Levin M, Frisch J: Plasma cell granulaoma of periodontal tissues. Periodontics 6: 272–276, 1968.
3. Albright RL, Finkelman A, Doner JM, Beaubien R: Multiple myeloma with manifestation of a maxillary bony lesion and plasmacytoma. Oral Surg 26: 167–172, 1968.
4. Warson R, Preis F: A nonexophytic plasma-cell granuloma of mandible. Oral Surg 28: 791–796, 1969.
5. Loh HS: A retrospective evaluation of 23 reported cases of solitary plasmacytoma of the mandible, with additional case report. British J Oral and Maxillofacial Surg 22: 216–224, 1984.
6. Matinelli C, Rulli MA: Primary Plasmacytoma of soft tissue (gingiva). Oral Surg 25: 607–609, 1968.
7. Koop SH, Carely RB: Extramedullary plasmacytoma of salivary glands. Laryngoscope 76: 1971–1973, 1966.
8. Pascoe HR, Dorfman RF: Extramedullary plasmacytoma of salivary glands. Am J Cli Pathol 51: 501–507, 1969.
9. Gaston EA, Dollinger MR, Strong EW, Hajdu SI: Primary plasmacytoma of lymph nodes. Lymphology 1: 7–15, 1969.
10. Mattehews JR, Baus MK: Plasma cell lesions within the oral tissue Immunoperoxidase staining of routinely fixed and processed tissue. Oral Surg 54: 414–423, 1982.
11. Sternberger LA: Immunocytochemistry. John Wiley, New York, 3rd ed, 1974.
12. Bataille R, Sany J: Solitary myeloma: Clinical and prognostic features of a review of 114 cases. Cancer 48: 845–851, 1982.
13. Mill NB, Griffith R: The role of radiation therapy in the management of plasma cell tumors. Cancer 45: 647–652, 1980.
14. Woodruff RK, Whittle JN, Malpas JS: Solitary plasmactoma II: Solitary plasmcytoma of bone. Cancer 43: 2344–2347, 1979.
15. Tong D, Griffin TW, Laramore GE, Kurtz JM, Russell AH, Grondine MT, Herron T, Blasbo JC, Tesh DW: Solitary plasmacytoma of bone and soft tissues. Radiology 135: 195–198, 1980.
16. Kotner LM, Wang CC: Plasmacytoma of the upper air and food passages. Cancer 30: 414–418, 1972.
17. Kalderon AE, Bogaars HA, Diamond I, Cummings FJ, Kaplan SR, Calabresi P: Ultrastructure of myeloma cells in a case with crystal-cryoglobulinemia. Cancer 39: 1475–1481, 1977.
18. Sorenson GD: Electron microscopic observations of bone marrow from patients with multiple myeloma. Lab Invest 13: 196–313, 1964.
19. Maldonado JE, Brown AL, Bayard ED, Pease GL: Ultrastructure of myeloma cell. Cancer 19: 1613–1627, 1966.
20. Azar HA, Zaino EC, Pham TD, Yannopoulos K: "Nonsecretory" plasma cell myeloma: observations on seven cases with electron microscopic studies. Am J Clin Pathol 58: 618–629, 1972.
21. Smetana K, Cyorkey F, Cyorkey P, Busch H: Ultrastructural studies on human myeloma plasmacytes. Cancer Res 33: 2300–2309, 1973.
22. Oikawa K: Electron microscopic obser-

vations of inclusion bodies in plasma cells of multiple myeloma and Waldenstrom's macroglobulinemia. Tohoku J Exp Med 117: 257–282, 1975.
23. Brunning RD, Parkin J: Intranuclear inclusions in plasma cells and lymphocytes from patients with monoclonal gammopathies. Am J Clin Path 66: 10–21, 1976.
24. Taylor CR, Russel R, Chandor S: An immunohistochemical study of multiple myeloma and related conditions, using an immunoperoxidase method. Am J Clin Pathol 70: 612–622, 1978.

IMMUNOHISTOCHEMICAL DETECTION AND ULTRASTRUCTURAL FEATURES OF SOLITARY PLASMACYTOMA OF MAXILLARY BONE —— CASE PRESENTATION

YEONG-LEI HUANG, LI-MIN LIN, CHUNG-HO CHEN
CHENG-CHUNG LIN, YAT-HANG YAN

Plasma cell neoplasm have been classified as multiple myeloma, solitary plasmacytoma and extramedullary plasmacytoma. The solitary plasmacytoma of maxilla is a rare condition. It is a single focus of myelomatous tissue with no dissemination to other parts of the skeleton. This paper presents a case of solitary plasmacytoma in the maxillary bone. Roentogenographic examination revealed a cystic-like osteolytic lesion over the left posterior portion of maxillary bone, invading maxillary sinus. The CT scan showed the tumor mass occupied the left maxillary sinus and lateral wall of the nasal cavity, protruding to the buccal side of the left face. The tumor cells were composed of densely packed round and polygonal cell structures which were scattered in relatively sprase stoma. The neoplastic cells have large, single eccentric nucleus, resembling typical plasma cells. The ultrastructural studies of tumor cells revealed numerous endoplasmic reticulum. There were arranged in a lamellar pattern, large numbers of mitochondria in perinuclear distribution and prominent Golgi complex. Nuclei showed patch condensation of chromatin and large nucleoli. The monoclonal staining (anti-Kappa and anti-IgG) of tumor cells by the peroxidase anti-peroxidase immunohistochemical techique, proved that the plasma cell lesion is neoplastic in nature. The tumor mass was eradicated by total resection, followed by X-ray radiation (4200 RADS). The patient is in good physical condition. There has been no clinical evidence of recurrence three years after surgery.

School of Dentistry, Kaohsiung Medical College, Kaohsiung 80708, Taiwan, Republic of China.