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ORIGINAL ARTICLE

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High-grade carcinoma component in epithelial-myoepithelial carcinoma of salivary glands clinicopathological, immunohistochemical and flow-cytometric study of three cases

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Abstract Three cases of epithelial-myoepithelial carcinoma (EMC) with coexisting areas of high grade carcinoma are reported. In two of the cases there was a previous recurrence, and in all three patients there had been a sudden increase in size before final surgery. The typical ductal and myoepithelial components of EMC showed the usual biphasic pattern and the expected immunophenotypes, with expression of wide spectrum cytokeratins, Cam 5.2 and EMA in the ductal part, and muscle-specific actin, smooth muscle actin, S-100 protein, vimentin and cytokeratins in the myoepithelial component. These areas also had a low mitotic count and low proliferation rate as measured by immunohistochemistry and by flow cytometry. Conversely, areas of high-grade tumour had the features of a large cell carcinoma, with focal mucin secretion in two cases. This high-grade component showed an epithelial immunophenotype in two cases, and was negative for all tested markers in the third one. The mitotic counts and the proliferation rates were much higher in these anaplastic areas. One of the patients died 3 months after treatment; another developed lymph node metastases 1 year later and was alive after 6 years of follow-up. The third patient was alive without evidence of disease 7 months after wide surgical resection of the tumour. The possibility of anaplastic transformation in EMC makes thorough sampling mandatory in this type of neoplasm.

Key words Epithelial myoepithelial carcinoma · Salivary gland tumours · Immunohistochemistry · Flow cytometry · MIB-1 (Ki 67) · Proliferative markers

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Introduction

Epithelial-myoepithelial carcinoma of intercalated duct origin (EMC) is an uncommon low-grade salivary gland neoplasm that accounts for under 1% of salivary gland tumours [25, 28]. The entity was first described in Germany by Donath et al. in 1972 [10]. In decades since then, EMC has gained acceptance in the English and French literature as different series have been reported [3, 8, 19]; it was recognized as a distinct pathologic entity in the 1991 World Health Organization (WHO) and AFIP classifications of salivary gland tumours [12, 28]. Before its truly malignant nature had been recognized, this neoplasm had been illustrated in the 1972 WHO classification of salivary gland tumours [34] and the AFIP Tumour Fascicle [33] as a "clear cell monomorphic adenoma". Although histologically EMC exhibits a high degree of differentiation, it has been shown to be a low-grade carcinoma because of its infiltrative and destructive growth tendency, the presence of foci of necrosis, frequent recurrences, and perineural involvement with distant metastases [10].

Despite its high degree of cell differentiation and its low-grade aggressiveness, areas with cellular pleomorphism and nuclear atypia have been observed in some "bona fide" EMC [13, 30]. Also, the very rare association of EMC with adenoid cystic carcinoma, which is a high-grade tumour, has been described [7]. However, the association of EMC of salivary glands with undifferentiated high-grade carcinoma has not yet been reported.

We report three cases of EMC coexisting with undifferentiated high-grade carcinoma. Two of them originated in the parotid gland and one from minor salivary

glands of the palate. In these three tumours, the low- and high-grade components have been compared for immunophenotype, proliferative activity and ploidy.

Materials and methods

The medical records of the three patients affected by these neoplasms were reviewed. Histopathological studies were carried out on haematoxylin and eosin-stained slides with and without formalin fixation. Representative paraffin blocks were selected for histochemical and immunohistochemical procedures. Special stains included the periodic acid-Schiff stain (PAS), with and without diastase digestion, alcian blue and mucicarmine stain. Immunohistochemical studies were performed with the avidin–biotin–peroxidase complex technique, and the following primary antibodies were used: cytokeratin cocktail, low-molecular-weight cytokeratin (CAM 5.2), epithelial membrane antigen (EMA), carcinoembryonic antigen (CEA), muscle-specific actin (MSA), smooth muscle actin (SMA), S-100 protein, vimentin, desmin and glial fibrillary acidic protein (GFAP). A summary of the source, type and working dilution of each of these antibodies is given in Table 1. The immunoreactivity was called positive when all or almost all of the tumour cells showed strong positivity. When only a few scattered tumour cells or a small group of tumour cells were positive, they were labelled as focally positive.

Proliferative activity was studied immunohistochemically by means of the MIB-1 (Ki-67) antibody. Antigenic retrieval was performed with heat pre-incubation in citrate buffer with a steam sterilizer for 1 h prior to primary antibody incubation. The sections stained with MIB-1 were evaluated at high-power field magnification (HPF) (×400), using a microscopic grid. A median number of 3000 cells of every type (ductal, myoepithelial or high-grade neoplastic cells) were counted. Tumour cells were considered positive for MIB-1 when any staining of the nuclei, regardless of the staining intensity, was found.

Flow-cytometric analysis of the nuclear DNA content was performed in each case by studying the typical EMC areas and the high-grade tumour separately. The technique was applied on 50-µm-thick sections obtained from the paraffin blocks following Hedley's method [16]. Propidium iodide was used as fluorochrome. Samples were analysed by an Epics Profile II flow cytometer (Coulter, Healeh, Fla.), and single-parameter histograms used for tumour evaluation were analysed with the multicycle Software (Phoenix Flow Systems, San Diego, Calif.). They were classified according to the Guidelines of the Committee on Nomenclature of the Society for Analytical Cytology [29]. Ploidy, S-phase fraction (SPF) and proliferative index (PI; SPF+G2M fraction) were recorded for each case. The coefficient of variation was evaluated in all samples, and the mean value was 5.58 (range 3.6–8.3).

Table 1 Panel of Antibodies used in the study (CAM low-molecular-wight cytokeratin, *CEA* carcinoembryonic antigen, *EMA* epithelial membrane antigen, *GFAP* glial fibrillary acidic protein, *MSA* muscle-specific actin, *SMA* smooth muscle actin; *M* monoclonal, *P* polyclonal)

Antibody	Type	Source	Dilution
Cytokeratins	P	Dako (Glostrup, Denmark)	1:300
CAM 5.2	M	Becton-Dickinson (San Jose, Calif.)	1:100
EMA	M	Dako	1:200
CEA	M	Euro-Diagnostica (Apeldoorn, Holland)	1:50
MSA (HHF35)	M	Enzo Diagnostics (Syosset, N.Y.)	1:50
SMA	M	Dako	1:800
S-100 Protein	P	Dako	1:500
Vimentin	M	Dako	1:100
Desmin	M	Dako	1:100
GFAP	P	Dako	1:300
MIB-1	M	Biogenex (San Ramon,Calif.)	1:20

Results

The clinicopathological findings are summarised in Table 2.

Case 1

An 88-year-old woman presented with a tumour mass in the left parotid gland, which protruded widely in the ipsilateral hemifacial region. The CT scan demonstrated extension of the growth from the zygomatic arch and cheek to the retromandibular neck (Fig. 1). The tumour

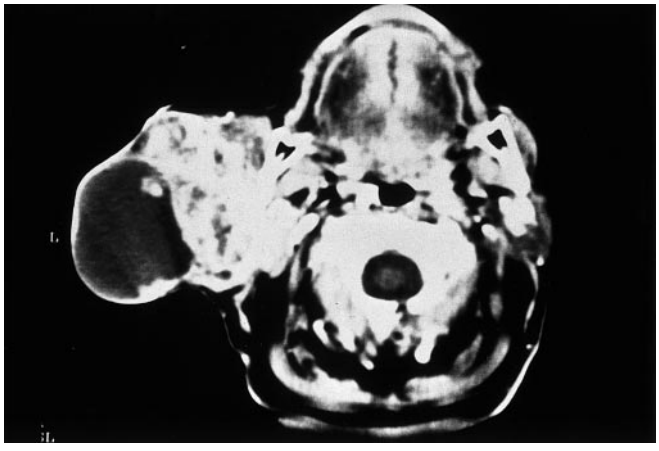


Fig. 1 Case 1. CT scan demonstrating growth of a cystic tumour beyond the left parotid gland

Table 2 Clinicopathological findings (*F* female, *M* male, *DOD* dead of disease, *NED* not evidence of disease)

Case no.	Age (years)	Sex	Location	Size (cm)	Presence of low-grade tumour (years)	High-grade neoplasia	Follow-up
1	88	F	Parotid gland	11×8x6	14	Adenocarcinoma	DOD 3 months
2	78	M	Parotid gland	4×3x2	4	Undifferentiated large cell carcinoma	NED 6 years
3	66	F	Palate	8×6x5.5	35	Adenocarcinoma	NED 7 months

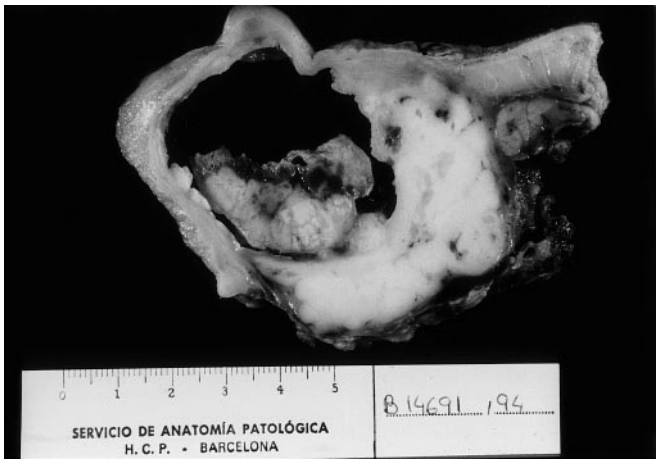


Fig. 2 Case 1. Parotid tumour attached to the skin, showing irregular margins and a large necrotic central area forming a cystic cavity

had started as a small preauricular nodule 14 years before and had grown slowly but relentlessly since then. An acceleration of growth had been noticed 2 months prior to hospital admission. The tumour was removed by extensive surgery. The surgical specimen included the whole parotid gland, the masseter muscle, periostium of the ascending mandibular ramus and the lower half of the lobule of the left ear. The tumour measured 11×8×6 cm, had irregular margins and showed large necrotic areas forming a cystic cavity (Fig. 2). In spite of treatment, the patient showed progressive general deterioration, and a metastatic hepatomegaly was demonstrated by echography. She died 3 months later.

Case 2

A 78-year-old man presented with a painless tumour in his right parotid region. The lump had been present for 2 years, with slow progressive growth. A fine-needle aspiration (FNA) cytology of the lesion showed clusters of cells with the appearance of epithelial cells, with clear cytoplasm and small uniform nuclei. There were also fragments of pale, homogeneous, acellular material, which were found either in isolation or surrounding cell clusters. These findings were interpreted as indicating an epithelial neoplasm of probable salivary gland origin. The patient refused surgery until 2 years later, when the size of the lesion suddenly increased. A total parotidectomy with facial nerve conservation was then performed. The surgical specimen showed a solid, well-circumscribed, round mass measuring 4×3×2 cm involving the deep lobe of the parotid gland. One year later, a right cervical lymph node metastasis was detected and treated by radical neck dissection plus radiotherapy. The patient is alive, without evidence of disease, 6 years after the first surgery.

Case 3

A 66-year-old woman had had a tumour removed from the hard palate at the age of 31. Two years later the tumour recurred, showing a very slow growth. The patient let it evolve without further treatment until 35 years after removal of the first tumour. At this time, the CT scan showed a destructive tumour infiltrating the right hard palate and the floor of the right maxillary sinus. It had also infiltrated the right pterygoid muscles and destroyed the right pterygoid apophysis. The tumour was solid, with heterogeneous density and some calcifications. A wide surgical resection of the tumour was performed, with removal of the anterior and medial maxillary walls and of the right hard palate. The tumour mass was irregular, had a firm consistency, and measured 8×6×5.5 cm. Seven months after surgery the patient is alive with no evidence of disease.

A summary of the immunohistochemical results is presented in Table 3, and the flow-cytometric results and those relating to the proliferation markers are shown in Table 4.

Case 1

The EMC component of the neoplasm was well differentiated and exhibited a multinodular growth pattern with

Table 3 Immunohistochemical results (*f* focal positivity, *n.e* non evaluable, *Myoep* myoepithelial, *ca* carcinoma)

Antibodies	Case 1			Case 2			Case 3		
	Ductal cell	Myoep cell	High-grade ca	Ductal cell	Myoep cell	High-grade ca	Ductal cell	Myoep cell	High-grade ca
Cytokeratins	+	+	+	+	+	–	+	+	+
CAM 5.2	+	+	+	+	f	–	+	f	+
EMA	+	–	+	+	–	–	+	–	f
CEA	+	–	+	f	–	–	–	–	f
MSA	–	+	–	–	+	–	–	+	–
SMA	–	+	f	–	+	–	–	+	–
S-100 Protein	–	+	–	–	+	–	–	+	f
Vimentin	–	+	–	–	+	–	–	f	–
Desmin	–	–	–	–	–	–	–	–	–
GFAP	–	–	–	–	–	–	–	f	–
MIB-1	0.4%	3%	30%	n.e.	n.e.	n.e.	0.3%	2%	12%

Table 4 Flow cytometric and proliferative marker results (*EMC* epithelial-myoepithelial carcinoma, *HGC* high grade carcinoma, *HPF* high power field [×400], *PI* proliferation index)

Histological areas	Case 1		Case 2		Case 3	
	EMC	HGC	EMC	HGC	EMC	HGC
Mitosis	0/10HPF	20/10HPF	0/10HPF	6/10HPF	0/10HPF	8/10HPF
Ploidy	Diploid	Diploid	Diploid	Diploid	Diploid	Diploid
S-Phase fraction (SPF)	6.2%	19.5%	9.1%	n.e	8.4%	9.8%
PI (SPF+G_2 M)	10.2%	24.8%	13.4%	n.e	12.7%	20.7%
MIB-1	Clear cells 3% Ductal cells 0.4%	30%	n.e	n.e	Clear cells 2% Ductal cells 0.3%	12%

islands of tumour separated by dense fibrous connective tissue septa (Fig. 3). Small ducts were lined with cuboidal epithelium and surrounded by clear cells overlying an external basement membrane, thus showing a biphasic pattern. The cuboidal cells had finely granular, dense eosinophilic cytoplasm and central or basally located, round nuclei. The clear cells were polyhedral with abundant cytoplasm, well-defined borders and slightly eccentric vesicular nuclei. The cytoplasm of the clear cells was PAS positive and diastase soluble, indicating the presence of glycogen. Some ductal lumina contained eosinophilic secretory material. Focally, cystic dilatations of the ducts were seen, along with some tumour protusions and discrete papillary proliferation of the two types of tumour cells. Extensive areas were composed exclusively of clear cells, devoid of ducts and sometimes forming solid neoplastic masses or a trabecular growth pattern with hyalinized fibroconnective tissue between solid cellular groups. These areas showed variable ischaemic necrosis. Mitotic figures were practically absent. The immunohistochemical studies revealed diffuse positivity of the ductal cells for broad-spectrum cytokeratins, CAM 5.2, EMA and CEA. The clear cells expressed the characteristic phenotype of the myoepithelial cells, being positive for MSA, SMA, S-100 protein, cytokeratins, CAM 5.2 and vimentin. The proliferative activity measured by the expression of MIB-1 antibody revealed positivity in 0.4% of the ductal cells and 3% of the clear cells. The flow-cytometric study, in these areas, showed a DNA diploid (euploid) population with an SPF of 6.2% and PI of 19.5%.

In continuity with the typical EMC we identified areas showing more active growth of the epithelial component, with increased atypia of the nuclei. These areas of activated epithelium formed solid nests surrounded by a layer of clear myoepithelial cells (Fig. 4). A different histological pattern was observed close to the areas previously described. This component formed solid groups of cells with central necrosis, occasionally of the comedo type, and contained large epithelial cells exhibiting prominent infiltrative growth with desmoplastic reaction (Fig. 5). These cells showed abundant clear or slightly eosinophilic cytoplasm with vesiculous and pleomorphic nuclei harbouring prominent nucleoli (Fig. 6). Focally, scarce lumina with mucous secretion positive for alcian blue, PAS and mucicarmine stains were seen. The mitotic count was high in this component, being 20 mitoses/10 HPF. Immunohistochemical studies demonstrated positivity of these cells for cytokeratins, CAM 5.2, EMA and CEA. The immunoreaction for SMA was focally positive in the periphery of the solid nests. The MIB-1 antibody was positive in 30% of the neoplastic cells of these high-grade areas. As in the low-grade areas, flow-cytometric analysis showed the presence of a DNA diploid (euploid) population. However, the SPF was considerably higher (19.5%) and the PI was 24.8%.

Case 2

Most of the tumour was a well-differentiated EMC with a bicellular pattern, presence of cuboidal cells forming ductal structures, and large areas of myoepithelial clear cells showing solid nests devoid of ducts in some fields. Ischaemic necrosis was focally present. The mitotic activity was practically absent in the well-differentiated component. The immunohistochemical profile of this component was similar to case 1. However, the clear cells showed only focal positivity for CAM 5.2. The flow-cytometric analysis showed a DNA diploid (euploid) population with an SPF of 9.1%.

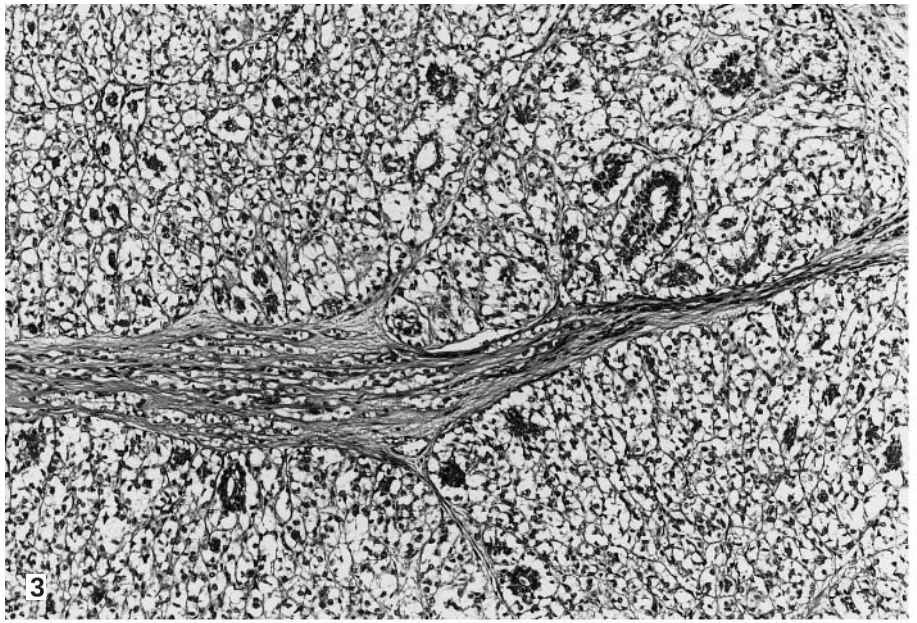
In continuity with the typical EMC, a proliferation of poorly differentiated cells, with abundant clear cytoplasm, pleomorphic nuclei, and prominent nucleoli was identified. This second tumour component grew in a solid, diffuse, infiltrative pattern having the histological features of an undifferentiated large cell carcinoma (Fig. 7).

Fig. 3 Case 1. Typical epithelial-myoepithelial carcinoma (EMC) component of the neoplasm. The tumour is composed of centrally placed small ducts lined with dark cuboidal epithelium and surrounded by abundant layers of clear cells. Fibrous septa are present. HE, ×100

Fig. 4 Case 1. Transition of EMC to a more active epithelial growth. The activated epithelial cells are surrounded by a layer of clear myoepithelial cells. HE, ×100

Fig. 5 Case 1. Groups of poorly differentiated epithelial cells (*left*), surrounded by a fibrous reaction, growing in the vicinity of typical EMC (*right*). HE, ×100

Fig. 6 Case 1. High-grade carcinoma component. The cells are large with vesicular and pleomorphic nuclei showing prominent nucleoli. HE, ×400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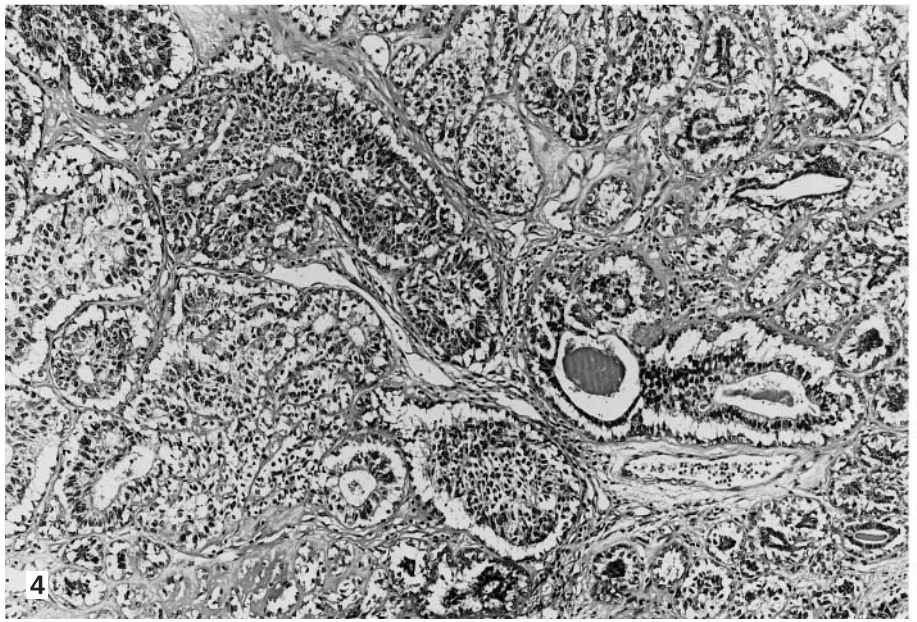
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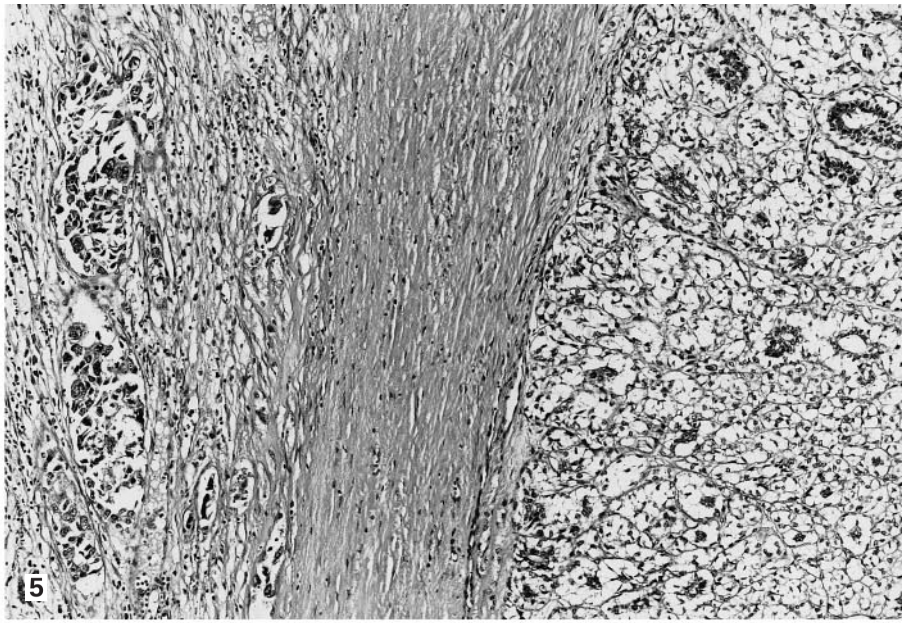
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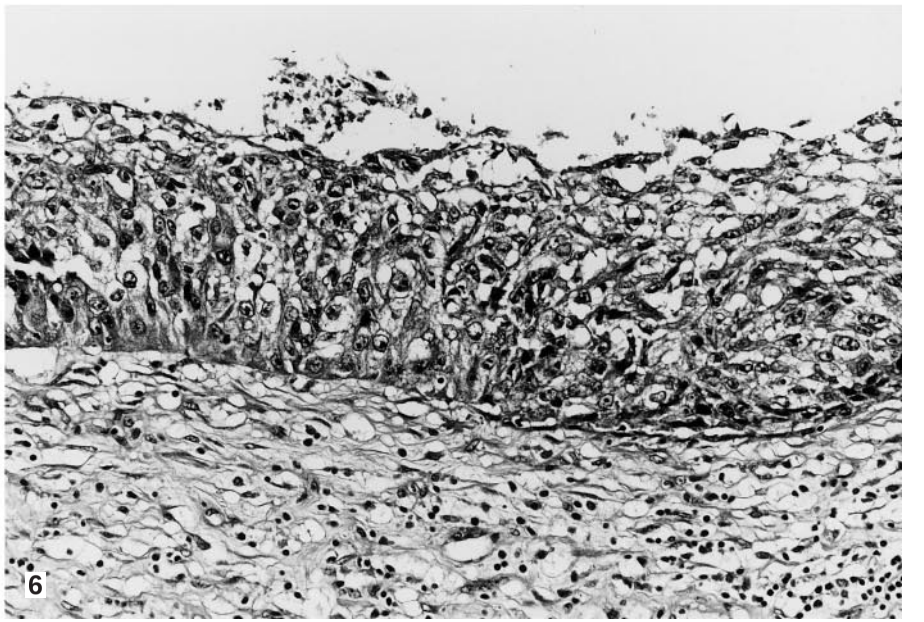
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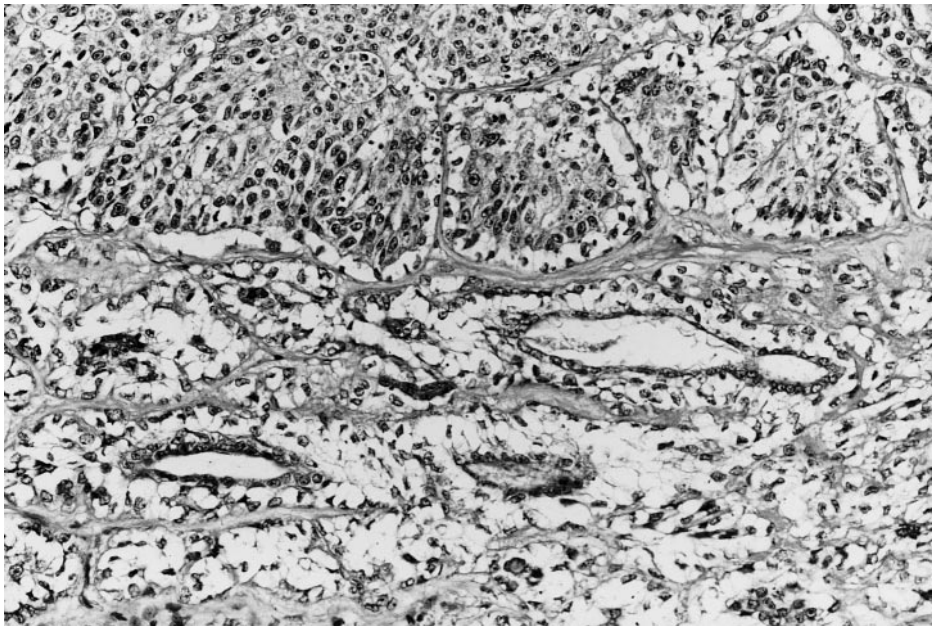

Fig. 7 Case 2. Typical EMC areas in continuity with a high-grade carcinoma composed of large cells with pleomorphic and atypical nuclei. HE, ×200

The mitotic count in these areas was 6/10 HPF. All the immunohistochemical markers proved negative in these areas. The flow-cytometric analysis of the high-grade areas was meaningless because of the low ratio of nuclei obtained. Immunohistochemical evaluation of the MIB-1 antibody was impractical.

The cervical lymph node metastasis detected in this patient 1 year later showed the pattern of a high-grade carcinoma, being identical to that observed in the parotid gland.

Case 3

In this case, the biphasic and well-differentiated areas composed of ductal and clear cells were similar to those observed in cases 1 and 2, and the mitotic count was also low. Cystic dilatations with encephaloid protusions and focal papillary proliferations were evident focally. Foci of squamous metaplasia among abundant clear cells were commonly seen (Fig. 8). Stromal microcalcifications were also present. The immunohistochemical studies demonstrated positivity of the ductal cells for cytokeratins, CAM 5.2 and EMA, whereas the clear cells were positive for cytokeratins, MSA, SMA, S-100 protein, and focally for CAM 5.2, vimentin and GFAP. The MIB-1 immunostaining was positive in 0.3% of the ductal cells and in 2% of the myoepithelial cells. The flow-cytometric profile of this component showed a DNA diploid (euploid) population with an SPF of 8.4% and PI of 12.7%

A second tumour component was seen close to the well-differentiated areas. It was composed of large cells with eosinophilic cytoplasm and pleomorphic nuclei, which formed some lumina (Fig. 9) with scarce and focally alcian-blue- and mucicarmine-positive secretions. Foci of necrosis were also seen in these high-grade areas. The mitotic count was 8/10 HPF. The immunohistochemical studies revealed positivity for cytokeratins, CAM 5.2 and, focally, for CEA and S-100 protein. MIB-1 was positive in 12% of the neoplastic cells in these areas. The flow-cytometric analysis showed a DNA diploid (euploid) population with an SPF of 9.8% and PI of 20.7%.

Discussion

EMC usually arises from the parotid gland, and more rarely from minor salivary glands, involving the maxillary antrum and palate [8, 14, 19]. This neoplasm exhibits a predilection for female subjects and for old age, with a peak incidence in the seventh decade [6, 8, 9, 15, 19], although cases in children have been reported [21].

EMC is a biphasic neoplasm composed of epithelial and myoepithelial cells presenting with a broad spectrum of histopathological appearances [2, 13]. The tumour usually shows a very typical growth pattern on low magnification, with lobular, advancing, margins and an unmistakable bicellular appearance formed by epithelial ductal cells, usually with an intercalated duct appearance, surrounded by a mantle of cells with optically clear cytoplasm and with a myoepithelial phenotype. Some areas with a predominance of clear cells and a paucity of epithelium-lined ducts and ductules are frequently observed. Other patterns described include the presence of spindle myoepithelial elements, cystic dilatation with encephaloid protusions and papillary proliferations in the biphasic areas and foci of squamous metaplasia [2, 4]. Interestingly, areas of dedifferentiation in recurrences and metastases have been described previously [30]. These areas contained pleomorphic spindle cells, prominent haemorrhage, necrosis and numerous mitotic figures, so that there is a correlation between these histological changes and a worse prognosis [30]. However,

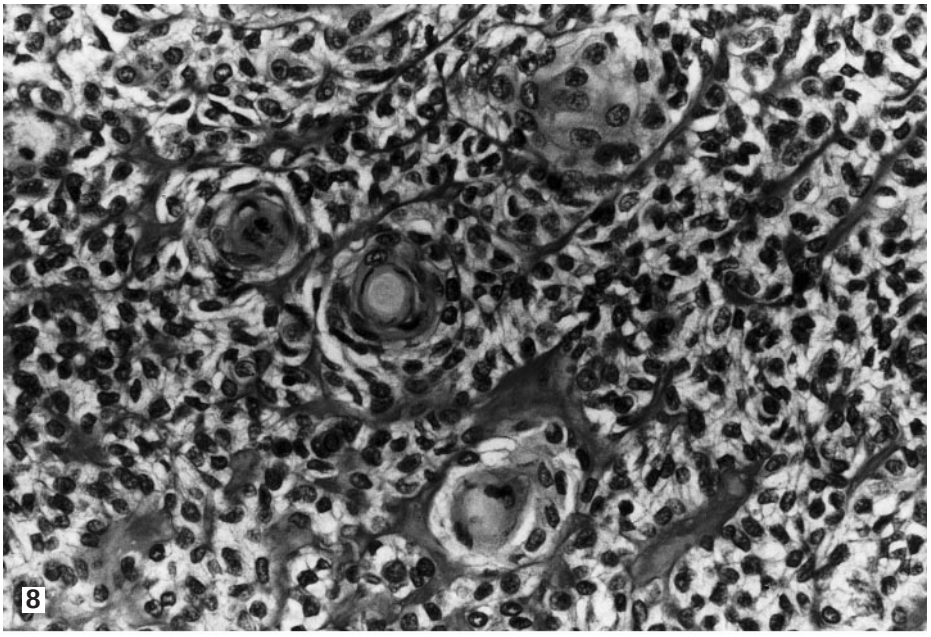

Fig. 8 Case 3. EMC pattern with clear cell predominance and foci of squamous metaplasia. HE, ×400

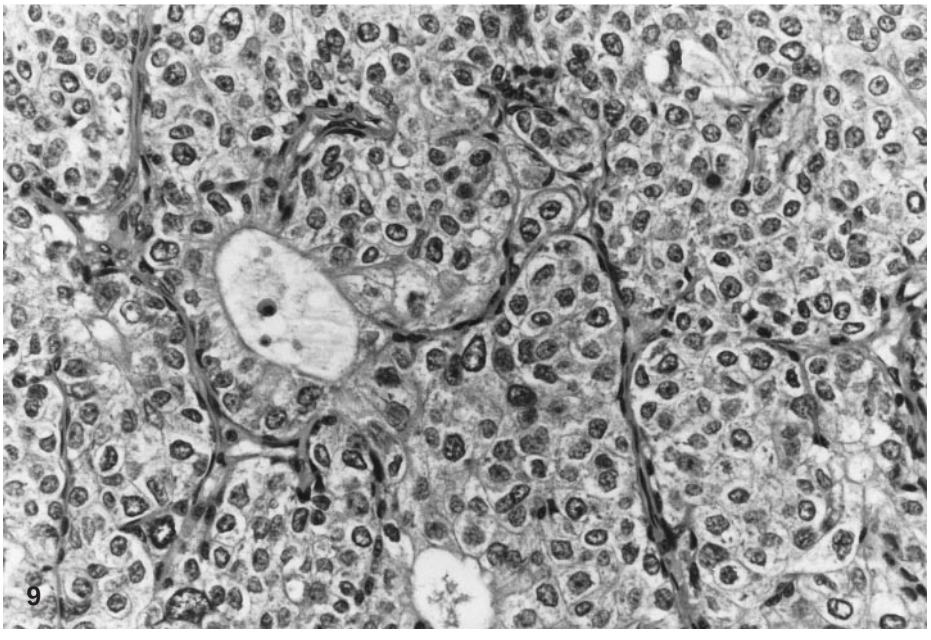

Fig. 9 Case 3. High-grade carcinoma component composed of large cells with vesicular nuclei and patent nucleoli. There is a tendency to glands formation. HE, ×400

except for the association with an adenoid-cystic carcinoma [7], no other different neoplasm has been described as associated with an EMC in the salivary glands.

In our three cases, extensive areas of typical EMC consisting of epithelial ductal and myoepithelial components were observed, and their origins were confirmed immunohistochemically. The three cases also harboured areas of a high-grade neoplastic component, which exhibited the morphological features of a high-grade adenocarcinoma in cases 1 and 3. Areas exhibiting active growth of the epithelial component with progressive cellular atypia, indicating transition from low-grade to high-grade carcinoma, were seen in case 1. The epithelial nature of the high-grade component was confirmed immunohistochemically; it showed a reactivity profile characteristically different from malignant myoepithelioma [1]. In case 1, the focal and peripheral positivity for SMA was attributed to the presence of stromal myofibroblasts and vessels. All the immunohistochemical studies turned out to be negative in the high-grade component of case 2, in spite of the typical immunohistochemical profile of the low-grade EMC component.

In these three cases, the tumours were known to have been present for several years before they began to increase in size and aggressiveness. The behaviour of the typical salivary gland EMC is that of a low-grade malignant neoplasm, with slow locally infiltrative growth and the possibility of recurrences after surgical excision. In rare cases it can give rise to remote metastases and even-

tually kill the patient [6, 8, 15, 19]. In attempts to predict the biological behaviour of salivary gland EMC, different variables have been studied, including tumour size, histopathological pattern, DNA analysis by image cytophotometry and proliferative activity measured by the Ki-67 antibody, but none has correlated with the biological course and prognosis of this neoplasm [5, 13, 15].The only morphological feature that has been found to correlate with prognosis is the presence of nuclear atypia in more than 20% of the tumour cells [13]. The evaluation of proliferative activity by Ki-67 analysis has been shown to correlate with clinicopathological factors and patient survival in several neoplastic conditions, including salivary gland neoplasms [17, 22]. Therefore, attention was directed towards establishing the pattern of proliferative activity in the different components of our three tumours.The anaplastic areas of our three cases exhibited higher proliferative activity than the typical EMC foci. Interestingly, the myoepithelial component of the latter had a higher mitotic count than the ductal one. This supports the suggestion that myoepithelial cells play an important part in the growth of EMC, being capable of multiplying and giving rise to other tumour cell types [5]. It is tempting to consider that the high-grade carcinoma component of our cases was derived from myoepithelial cells, although we have no convincing evidence to offer. The existence of intense proliferation correlated with aggressive behaviour of the neoplasm in two of our cases, mainly in case 1, in which the patient died soon after the initial diagnosis and had the highest proliferation rates. Nevertheless, the value of the assessment of proliferation in tumours with this high-grade component is still too limited, both by the small number of cases studied and by the brief follow-up, to allow definitive conclusions. Future studies should consider the relative frequency of the association between EMC and a high-grade carcinoma.

Neoplasms with characteristics similar to EMC have been reported in the breast, where they have been called adenomyoepitheliomas [20, 32]. This is a misnomer: they are low-grade malignant tumours and should also be called epithelial-myoepithelial carcinomas [26, 27]. These tumours, although usually behaving as benign entities, can show a spectrum of behaviour like that seen in the salivary glands, with local recurrences and axillary lymph node and distant metastasis [18, 20, 23, 32]. The presence of a high-grade carcinoma and a malignant myoepithelioma coexisting with an adenomyoepithelioma associated with a fatal outcome has been described before [11, 20, 24], and other malignant components have been reported [31]. It seems reasonable to consider that such undifferentiated components in both breast adenomyoepitheliomas and EMC represent a transformation of low-grade tumours into high-grade lesions, rather than a collision of two different neoplasms.

These three cases of EMC with a high-grade carcinoma component confirm that the presence of this high-grade component confers an increased malignant potential on EMC, as shown by the high proliferative capacity and aggressive behaviour in two of our cases. In recurrent and long-standing EMC with sudden increase in size thorough sampling and evaluation of the proliferation activity are recommended.

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References

1. Alos L, Cardesa A, Bombi JA, Mallofré C, Cuchi A, Traserra J (1996) Myoepithelial tumors of salivary glands: a clinicopathologic, immunohistochemical, ultrastructural and flow-cytometric study. Semin Diagn Pathol 13:138–147
2. Batsakis JG, El-Naggar AK, Luna MA (1992) Epithelial-myoepithelial carcinoma of salivary glands. Ann Otol Rhinol Laryngol 101:540–542
3. Brocheriou C, Auriol M, de Roquancourt A, Gaulard P, Fornes P (1991) Carcinome épithélial-myoépithélial des glandes salivaires. Étude de 15 observations et revue de la littérature. Ann Pathol 11:316–321
4. Cheung FMF, Hioe F, Fong JHB (1995) Histologic variant of the epithelial-myoepithelial carcinoma of the salivary gland: a case report. Head Neck 17:437–444
5. Cho KJ, El-Naggar AK, Ordoñez NG; Luna MA, Austin J, Batsakis JG (1995) Epithelial-myoepithelial carcinoma of salivary glands. A clinicopathologic, DNA flow cytometric and immunohistochemical study of Ki-67 and HER-2/neu oncogene. Am J Clin Pathol 103:432–437
6. Collina G, Gale N, Visona A, Betts M, Cenacchi V, Eusebi V (1991) Epithelial-myoepithelial carcinoma of the parotid gland: a clinico-pathologic and immunohistochemical study of seven cases. Tumori 77:257–263
7. Corio RL (1991) Epithelial-myoepithelial carcinoma. In: Ellis GL, Auclair PL, Gnepp DR (eds) Surgical pathology of the salivary glands. Saunders, Philadelphia, pp 412–421
8. Corio RL, Sciubba JJ, Brannon RB, Batsakis JG (1982) Epithelial-myoepithelial carcinoma of intercalated duct origin. Oral Surg Oral Med Oral Pathol 53:280–287
9. Daley TD, Wysocki GP, Smout MS, Slinger RP (1984) Epithelial-myoepithelial carcinoma of salivary glands. Oral Surg 57:512–519
10. Donath K, Seifert G, Schmitz R (1972) Zur Diagnose and Ultrastruktur des tubulären Speichelgangkarzinoms. Epithelial-myoepitheliales Schaltstückkarzinom. Virchows Arch [A] 356:16–31
11. Dorpe J van, De Pauw A, Moerman P (1998) Adenoid cystic carcinoma arising in an adenomyoepithelioma of the breast. Virchows Arch 432:119–122
12. Ellis GL, Auclair PL (1996) Tumours of the salivary glands. (Atlas of tumour pathology, 3rd ser, fasc 17) Armed Forces Institute of Pathology, Washington DC, pp 268–281
13. Fonseca I, Soares J (1993) Epithelial-myoepithelial carcinoma of the salivary glands. A study of 22 cases. Virchows Arch [A] 422:389–396
14. Hagiwara T, Yoshida H, Takeda Y (1995) Epithelial-myoepithelial carcinoma of a minor salivary gland of the palate. Int J Maxillofac Surg 24:160–161
15. Hamper K, Brügmann M, Koppermann R, Caselitz J, Arps H, Askensten U, Auer G, Seifert G (1989) Epithelial-myoepithelial duct carcinoma of salivary glands: a follow-up and cytophotometric study of 21 cases. J Oral Pathol Med 18:299–304
16. Hedley DW, Friedlander MI, Taylor LW (1983) Method for analysis of cellular DNA content of paraffin-embedded pathological material using flow cytometry. J Histochem Cytochem 31:1333–1335
17. Hellquist HB, Karlsson MG, Nilsson C (1994) Salivary duct carcinoma-a highly aggressive salivary gland tumour with overexpression of c-erbB-2. J Pathol 172:35–44

18. Loose JH, Patchevsky AS, Hollander IJ, Lavin LS, Cooper HS, Katz SM (1992) Adenomyoepithelioma of the breast: a spectrum of biologic behavior. Am J Surg Pathol 16:868–876
19. Luna MA, Ordoñez NG, Mackay B, Batsakis JG, Guillamondegui O (1985) Salivary epithelial-myoepithelial carcinomas of intercalated ducts: a clinical, electron microscopic and immunocytochemical study. Oral Surg Oral Med Oral Pathol 59:482–490
20. Michal M, Baumruk L, Burger J, Mañhalová M (1994) Adenomyoepithelioma of the breast with undifferentiated carcinoma component. Histopathology 24:274–276
21. Morinaga S, Hashimoto S, Tezuka F (1992) Epithelial-myoepithelial carcinoma of the parotid gland in a child. Acta Pathol Jpn 42:358–363
22. Murakami M, Ohtani I Hojo H, Wakasa H (1992) Immunohistochemical evaluation with Ki-67: an application to salivary gland tumors. J Laryngol Otol 106:35–38
23. Pauwels C, DePotter C (1994) Adenomyoepithelioma of the breast with features of malignancy. Histopathology 24:94–96
24. Rasbridge SA, Millis RR (1998) Adenomyoepithelioma of the breast with malignant features. Virchows Arch 432:123–130
25. Seifert G (1996) Spezielle pathologische Anatomie, 2nd edn, vol 1/I: Oralpathologie I: Pathologie der Speicheldrüsen. Springer, Berlin Heidelberg New York, pp 590–600
26. Seifert G (1997) Terminologie des epithel-myoepithelialen Karzinoms der Speicheldrüsen und des Adenomyoepithelioms der Brustdrüse. Onkologe 3:666–671
27. Seifert (1998) Are adenomyoepithelioma of the breast and epithelial-myoepithelial carcinoma of the salivary glands identical tumours? Virchows Arch 433:285–287
28. Seifert G, Sobin LH (1991) Histological typing of salivary gland tumors, 2nd edn. (World Health Organization International Histological Classification of Tumors) Springer, Berlin Heidelberg New York, pp 23–24
29. Shankey TV, Rabinovitch PS, Bagwell B (1993) Guideliness for implementation of clinical DNA cytometry. Cytometry 14:472–477
30. Simpson RHW, Clarke TJ, Sarsfield PTL, Gluckman PGC (1991) Epithelial-myoepithelial carcinoma of salivary glands. J Clin Pathol 44:419–423
31. Simpson RHW, Cope N, Skálová A, Michal M (1998) Malignant adenomyoepithelioma of the breast with mixed osteogenic, spindle cell, and carcinomatous differentiation. Am J Surg Pathol 22:631–635
32. Tavassoli FA (1991) Myoepithelial lesions of the breast: myoepitheliosis, adenomyoepithelioma and myoepithelial carcinoma. Am J Surg Pathol 15:554–568
33. Thackray AC, Lucas RB (1974) Tumours of the major salivary glands. (Atlas of tumour pathology, 2nd ser, fasc 10) Armed Forces Institute of Pathology, Washington DC, pp 62–63
34. Thackray AC, Sobin LH (1972) Histological typing of salivary gland tumours. (International histological classification of tumours, no 7) World Health Organization, Geneva