**Myoepithelial Carcinoma of the Salivary Glands**

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**Objective:** To analyze the importance of unique cytoarchitectural patterns and the immunohistochemical profile in the diagnosis of myoepithelial carcinomas.

**Design:** Retrospective case analysis.

**Setting:** Tertiary cancer center.

**Patients:** A total of 51 patients with myoepithelial-rich carcinomas diagnosed over a 14-year period were studied for demographic data and tumor histologic characteristics and biologic behavior.

**Main Outcome Measures:** We analyzed various histopathologic parameters and an immunohistochemical profile consisting of pan-cytokeratin (Pan-CK), epithelial membrane antigen (EMA), CD10, smooth-muscle actin (SMA), S-100 protein, p63, calponin, and carcinoembryonic antigen (CEA).

**Results:** The parotid gland (n=15) and the palate (n=15) were common sites involved. The cell types encountered were epithelioid, stellate, plasmacytoid, spindle, clear, and mixed with myxoid, hyaline, or myxohyaline stroma. Immunohistochemical analysis revealed vimentin (100%), CK (74%), EMA (27%), CD10 (62%), SMA (35%), S-100 protein (82%), p63 (28%), and calponin (98%) positivity and CEA (100%) negativity. Cervical node dissection was performed in 17 cases: 7 showed nodal metastasis, 2 with pure spindle-cell morphologic characteristics and 3 with spindle cells mixed with other cells. Distant metastasis was noted in 3 of these 7 cases: 2 of these 3 cases showed spindle-cell morphologic characteristics.

**Conclusions:** Myoepithelial carcinomas showed varied cell types and patterns leading to a wide range of differential diagnoses. Immunohistochemical analysis helped determine the diagnosis. Spindle morphologic characteristics were observed with nodal and distant metastasis.

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**Myoepithelial Tumors** of the salivary gland including myoepitheliomas (benign) and myoepithelial carcinomas (malignant) are a rare group of tumors. Although myoepitheliomas were first described as early as 1943 by Sheldon,¹ the best description of myoepithelial tumors was given in the landmark articles by Dardick et al² in 1989 and Dardick³ in 1995. Myoepithelial tumors have also been included as a separate entity in the second edition of the World Health Organization’s histological classification of salivary gland tumors (1991).⁴⁵

Owing to morphologic heterogeneity, myoepithelial carcinomas have been grouped together and classified under the categories of “malignant mixed tumor” and “carcinoma ex pleomorphic adenoma.”⁶ The currently accepted diagnostic criteria for myoepithelial carcinoma are exclusive myoepithelial differentiation (morphologic and immunohistochemical) and clear-cut tumor infiltration into adjacent salivary gland or other tissues.⁷ To accurately identify this condition, the diagnostician must be aware of the entire spectrum of the cytoarchitectural patterns and the immunohistochemical (IHC) profile. This prompted us to analyze retrospectively all the cases diagnosed as myoepithelial tumors, malignant mixed tumors, and tumors with a predominant myoepithelial component arising in major and minor salivary glands. Those cases that fulfilled the criteria for myoepithelial carcinoma were included in the study. We present herein 51 cases of myoepithelial carcinoma of salivary gland origin and discuss their clinicopathologic features with particular emphasis on their morphologic spectrum.

**METHODS**

The present study includes 51 cases of myoepithelial carcinoma diagnosed in the Department of Surgical Pathology, Tata Memorial Hos-
hospital, Mumbai, India, over a period of 14 years (1992-2005). Exclusive myoepithelial differentiation (morphologic and IHC) and clear-cut tumor infiltration into adjacent salivary gland or other tissues were used as criteria for selection of cases. The clinical data and follow-up information were obtained from the medical records or the contributing pathologists.

The biopsy or other specimens received were fixed in 10% formalin. The tissue was embedded in paraffin, and 4-µm sections were examined after staining with hematoxylin-eosin. Hematoxylin-eosin–stained slides of all the tumors were viewed to assess a variety of histopathologic parameters, including cell type, growth pattern and architecture, pattern of infiltration, stroma (amount and type), nuclear atypia, mitosis, necrosis, perineural invasion, margin or bone involvement, and evidence of preexisting benign tumor.

We used the classification scheme for the cell types proposed by Dardick et al, Dardick, and Savaera et al. When more than 75% of the cells within the tumor were a single cell type, the tumor was classified as that cell type, whereas, when 2 or more cell types predominated, the tumor was designated as mixed cell type. The tumors were classified as high, intermediate, or low grade based on the degree of nuclear atypia and the presence or absence of necrosis and mitosis. High-grade tumors showed marked nuclear pleomorphism, chromatid clumping, prominent nucleoli, irregular nuclear membranes, and/or large areas of necrosis (>50% of the area) and mitosis (count, >4 to 10 per 10 high-power fields [hpf]). Presence of greater than 50% necrosis alone was considered an indicator of high-grade tumor. Intermediate-grade tumors showed moderate nuclear atypia with focal areas of necrosis and a mitotic count lower than 4 per 10 hpf, whereas low-grade carcinomas showed only mild nuclear atypia. Necrosis and mitosis were absent in low-grade tumors.

We performed IHC analysis on available additional sections in 50 cases. Before we incubated with antibodies, we submitted sections to antigen retrieval, using in most cases heat-induced epitope retrieval (20 minutes at 100°C in 10mM citrate buffer, pH 6.0) and/or trypsin digestion (0.1% in 0.2% calcium chloride solution, 10 minutes at 37°C). The sections were subjected to immunostaining with a labeled streptavidin–biotin complex. The signal was detected with 3,3-diaminobenzidine substrate. Biotin blocking agent was included only with few antibodies where required.

The following panel of antibodies was used: vimentin (monoclonal clone V9 [1:50]) (Dako, Glostrup, Denmark), pancytokeratin (Pan-CK) (monoclonal clone MNF116 [1:1000]) (Dako), epithelial membrane antigen (EMA) (monoclonal clone E20 [1:100]) (Dako), S-100 protein (polyclonal [1:300]) (Dako), smooth-muscle actin (SMA) (monoclonal clone 1A4 [1:200]) (Dako), calponin (monoclonal clone CALP [1:50]) (Dako), p63 (monoclonal clone 4A4 [1:50]) (Dako), CD10 (monoclonal clone 56C6 [1:40]) (NovoCastra, Leica Biosystems Nussloch GmbH, Nussloch, Germany), Ki-67 antigen (monoclonal clone Ki-55 [1:50]) (Dako), carcinoembryonic antigen (CEA) (polyclonal [1:400]) (Dako), and CD117 (C-kit) (polyclonal [1:100]) (Dako). Only 1 CK antibody analysis could be performed owing to financial constraints and lack of availability of material in referral and retrospective cases. For negative controls, we used a section from the same block from which the testing specimen was taken but added no antibody.

Tissue was available for electron microscopy (EM) in 2 cases. After fixing the specimens in 2.5% glutaraldehyde at 4°C for 2 hours, we then fixed them in 1% osmium tetroxide for 2 hours, dehydrated them through an ethanol series, transferred them to propylene oxide, and embedded them in epoxy resin. Ultrathin sections were cut on a Leica Ultramicrotome (Leica Biosystems) with a diamond knife. These sections were stained with uranyl acetate and lead acetate and examined under a Zeiss electron microscope (EM109: Carl Zeiss AG, Oberkochen, Germany).

We used χ² analysis to evaluate the influence of various morphologic features on prognosis.

RESULTS

CLINICAL DATA

The total number of salivary gland tumors diagnosed in the institute from 1991 to 2005 was 1243, of which 51 were classified as myoepithelial carcinomas (incidence, 4%). The clinical features of the 51 cases of myoepithelial carcinomas are summarized in Table 1 and Table 2. The cases included 30 men and 21 women, ranging in age from 14 to 70 years. Most of the patients were in their third to fifth decades of life (32 of 51, 63%). For most patients, the primary complaint was a painless mass. The duration of the symptoms ranged from 2 months to 6 years. Four patients with parotid tumors of 6 years’ duration had noted a recent rapid increase in size. Minor salivary gland involvement (n=36) was more common than major gland involvement (n=15). The parotid gland and palate were the most common primary sites (n=15 each). The other sites included buccal mucosa (n=7), nasal cavity (n=5), maxilla (n=3), lower alveolus (n=3), tongue (n=2), and the floor of the mouth (n=1).

PATHOLOGIC FEATURES: MACROSCOPIC FINDINGS

A wide range in tumor size was observed varying from 1 to 20 cm, with maximum number of tumors in the range of 1 to 5 cm (n=38). The mean tumor size was 4 cm. The tumors arising in the parotid gland were generally firm, solid, partially or completely encapsulated with nodular contours. At all other sites the tumors were unencapsulated. In some tumors, gelatinous zones were predominant. Necrosis was noted in 10 tumors grossly, whereas cystic change was observed in 6 cases.

PATHOLOGIC FEATURES: HISTOPATHOLOGIC FINDINGS

Cell Types

The tumor cells showed a wide morphologic variation.

Epithelioid. Epithelioid cells were the predominant cell type in 29% of tumors (15 of 51). The cells were polygonal and had central nuclei with coarse chromatin, prominent nucleoli, and pale eosinophilic cytoplasm. They also had ill-defined cell borders, were loosely cohesive (Figure 1A), and were arranged in sheets or trabeculae associated with varying amounts of hyaline or myxoid stroma. True glands with lumina were absent.

Plasmacytoid. Seven of the 51 tumors showed predominant plasmacytoid cell morphologic characteristics (14%) mimicking plasma cells (Figure 1B). All of the tumors showed hyaline stroma in varying amounts.

Spindle. Spindle-cell morphologic characteristics were seen in 12% of tumors (6 of 51). The cells were spindle shaped...
with centrally placed elongated nuclei. An interlacing fascicular pattern of arrangement of tumor cells was noted in all cases (Figure 1C). Stroma was absent in 5 of the 6 cases.

Clear. Three tumors were composed almost entirely of epithelioid cells with abundant clear cytoplasm (6%) (Figure 1D). The accompanying myxoid or hyaline stroma divided the tumor cells into small nests or cords.

Stellate. Eight of the 51 tumors showed unique cell morphologic characteristics (16%): the tumor cells were ovoid to short spindly with centrally placed nuclei, a moder-
ate amount of cytoplasm, and indistinct cell borders. The cells were arranged in a diffuse, sheetlike pattern (Figure 1E).

Mixed. In 12 of the 51 tumors (24%), a combination of 2 or more cell types (each making up more than 25% of the total number of cells) was noted; these tumors were classified as mixed-cell type. Epithelioid and plasmacytoid cells was noted in 6 cases, while epithelioid and clear cells were seen in 5 cases. Plasmacytoid with clear cells were seen in 1 case. All tumors of the spindle-cell type showed pure spindle cell morphologic characteristics.

Patterns of Infiltration

The tumors arising in the parotid gland showed partial or complete encapsulation microscopically. These tumors infiltrated through the capsule into the surrounding normal salivary gland or adjacent adipose or muscular tissue, with the extent of invasion varying from case to case. In contrast, assessment of invasion among tumors arising in minor salivary glands, especially the palate, was difficult owing to the unencapsulated nature of the tumors. In these cases, extension into surrounding bone, fat, or muscle was considered to be invasion. Tumors with multinodular architecture (both parotid and nonparotid tumors) showed a pattern of infiltration in the form of smaller nodules than those seen in the center of the tumor (Figure 2A). Cordlike infiltration was noted with tumors showing a diffuse pattern (n = 26) as well as those with a cordlike growth pattern. One case with a diffuse pattern of growth showed a nodular pattern of infiltration. Fascicular infiltration of tumor cells was observed with spindle-cell myoepithelial carcinomas. The cordlike pattern of infiltration (Figure 2B) was the most commonly seen (32 of 51, 63%), followed by the nodular pattern.

Growth Patterns and Architecture

A variety of growth patterns was observed, with a diffuse sheetlike arrangement of tumor cells being the most common (27 of 51, 53%), followed by a multinodular growth pattern (11 of 51, 22%). In 6 cases, the tumor cells showed a cordlike pattern made up of cords of cells separated by abundant stroma (12%). A compact fasciulated growth pattern was noted in all the spindle-cell type myoepithelial carcinomas (6 of 51, 12%). Stroma was absent in 5 cases of this pattern. In 1 case of a parotid tumor, a cribriform architecture was seen locally.

Table 2. Myoepithelial Carcinoma Tumor Sites and Clinical Parameters

<table>
<thead>
<tr>
<th>Site</th>
<th>Cases, No. (%)</th>
<th>Sex, No.</th>
<th>Age, Mean (Range), y</th>
<th>Size, Mean (Range), cm</th>
<th>Recurrence, No.</th>
<th>Metastasis, No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parotid</td>
<td>15 (29)</td>
<td>M 9</td>
<td>37.7 (16-65)</td>
<td>6.4 (2.2-20.0)</td>
<td>7</td>
<td>2</td>
</tr>
<tr>
<td>Palate</td>
<td>15 (29)</td>
<td>F 6</td>
<td>41.1 (18-66)</td>
<td>3.3 (1.0-7.0)</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>Buccal mucosa</td>
<td>7 (14)</td>
<td>M 5</td>
<td>42.3 (26-70)</td>
<td>4.1 (2.0-10.0)</td>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td>Nasal mass</td>
<td>5 (10)</td>
<td>F 3</td>
<td>45.4 (24-64)</td>
<td>4.3 (4.0-4.5)</td>
<td>2</td>
<td>0</td>
</tr>
<tr>
<td>Alveolus</td>
<td>3 (6)</td>
<td>M 2</td>
<td>21.3 (14-24)</td>
<td>4.3 (4.0-5.0)</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Maxilla</td>
<td>3 (6)</td>
<td>F 3</td>
<td>43.3 (24-70)</td>
<td>2.5 (2.0-3.0)</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Tongue</td>
<td>2 (6)</td>
<td>M 1</td>
<td>37 (32-42)</td>
<td>2.2 (2.0-2.5)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Floor of mouth</td>
<td>1 (2)</td>
<td>M 0</td>
<td>40 (40)</td>
<td>5.0 (5.0)</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Total</td>
<td>51 (100)</td>
<td>M 30</td>
<td>38.5 (14-70)</td>
<td>4 (1-20)</td>
<td>18</td>
<td>7</td>
</tr>
</tbody>
</table>

Stroma

Tumor-related matrix formed a distinct component in 46 cases. Hyaline stroma was the most commonly observed (26 of 51, 51%), followed by myxoid stroma (12 of 51, 24%). The myxoid stroma appeared bluish-gray, while the hyaline component was eosinophilic and fibrillar (Figure 1F). Mixed quality or myxohyaline stroma was seen in 8 of 51 cases (16%). Stroma was absent in 5 of 6 cases with spindle-cell morphologic characteristics.

Grading

Seven of the 51 tumors were classified as high grade (14%), 19 as intermediate grade (37%), and 25 as low grade (49%). High-grade tumors showed marked nuclear pleomorphism (Figure 2C) and/or large areas of necrosis and mitosis (count, >4 to 10 per 10 hpf) (Figure 2D). The presence of greater than 50% necrosis alone was considered an indicator of a high-grade tumor.

Associated Benign Tumor

Of the 51 tumors, 39 were de novo (77%), whereas the remaining 12 myoepithelial carcinomas showed unequivocal histologic evidence of an underlying benign tumor (24%). Nine of these 12 were conventional pleomorphic adenomas (labeled as carcinoma ex pleomorphic adenoma), and the other 3 were epithelioid (n = 2) and clear-cell (n = 1) myoepitheliomas. Of the 12 tumors associated with a preexisting benign tumor, 6 were parotid, and 6 were palatal. Of these 12 cases, 3 were high-grade, 6 were intermediate-grade, and 3 were low-grade carcinomas.

Other Features

Tumor metaplastic changes in the form of squamous metaplasia were seen in 3 cases. Chondroid or sebaceous metaplasia was not seen. Four of the 51 cases showed perineural invasion (8%), whereas angiomylymphatic invasion was seen in 2 cases. The palatal and maxillary sinuses...
Figure 1. Various architectural and cytologic patterns of myoepithelial carcinomas of the salivary glands (hematoxylin-eosin for all panels). A, Epithelioid-cell type (original magnification ×200). B, Plasmacytoid-cell type with myxoid stroma (original magnification ×400). C, Spindle-cell type (original magnification ×100). D, Clear-cell type (original magnification ×400). E, Stellate-cell type (original magnification ×200). F, Note the distinct eosinophilic hyaline stroma amidst tumor cells (original magnification ×200).
neoplasms demonstrated bone invasion in 6 of 25 cases. Mandibular bone invasion was not seen. Soft-tissue and/or mucosal resection margin involvement was seen in 5 of 44 cases in which major surgery was performed.

A few tumors had unusual morphologic characteristics such as extreme plasmacytoid differentiation, with cells showing prominent nucleoli and no stroma (resembling melanoma morphologically). Another case showed prominent rhabdoid differentiation resembling rhabdomyosarcoma. All of these cases (8 of 50) could only be resolved after IHC analysis.

**IHC PROFILE**

Immunohistochemical analysis was performed in 50 of the 51 cases, and the results of IHC staining are listed in **Table 3**. The IHC profile of myoepithelial carcinomas did not show any predilection with respect to different cell types. Vimentin (50 of 50, 100%) and calponin (49 of 50, 98%) findings were consistently positive in almost all of the tumors studied, followed by S-100 protein, which was immunoreactive in 41 of 50 tumors (82%) (Figure 3). Immunoreactivity for SMA was seen in 35% of cases (17 of 49) and was generally limited and weaker that seen with calponin. Pan-CK reactivity was noted in 74% of tumors (37 of 50) but was consistently absent in spindle-cell type tumors. All of the findings in tumors tested for CEA (n=42) were negative. Membranous staining for EMA was found.

**Table 3. Findings of IHC Analysis**

<table>
<thead>
<tr>
<th>IHC Test Substance</th>
<th>Positive Findings, No. (%) of Cases</th>
<th>Total, No. of Cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vimentin</td>
<td>50 (100)</td>
<td>50</td>
</tr>
<tr>
<td>Calponin</td>
<td>49 (98)</td>
<td>50</td>
</tr>
<tr>
<td>S-100 protein</td>
<td>41 (82)</td>
<td>50</td>
</tr>
<tr>
<td>CD10</td>
<td>31 (62)</td>
<td>50</td>
</tr>
<tr>
<td>SMA</td>
<td>17 (35)</td>
<td>49</td>
</tr>
<tr>
<td>p63</td>
<td>14 (28)</td>
<td>50</td>
</tr>
<tr>
<td>EMA</td>
<td>13 (27)</td>
<td>49</td>
</tr>
<tr>
<td>Ki-67</td>
<td>6 (12)</td>
<td>50</td>
</tr>
<tr>
<td>C-kit (CD117)</td>
<td>3 (6)</td>
<td>50</td>
</tr>
<tr>
<td>CEA</td>
<td>0</td>
<td>50</td>
</tr>
<tr>
<td>Vimentin + S-100 protein + calponin</td>
<td>40 (80)</td>
<td>50</td>
</tr>
<tr>
<td>S-100 protein + calponin + CD10</td>
<td>25 (50)</td>
<td>50</td>
</tr>
<tr>
<td>Calponin + CD10 + p63</td>
<td>10 (20)</td>
<td>50</td>
</tr>
</tbody>
</table>

Abbreviations: CEA, carcinoembryonic antigen; EMA, epithelial membrane antigen; IHC, immunohistochemical; SMA, smooth-muscle actin.
We found that CD10 and p63 were immunoreactive in 62% (31 of 50) and 28% (14 of 50) of tumors, respectively. In a few cases in which the morphologic characteristics were unusual (8 of 50), the tumor cells failed to show immunoreactivity to S-100 protein, SMA, and CK, and hence they were misdiagnosed initially as melanoma, rhabdomyosarcoma, schwannoma, and others, depending on the cytomorphologic characteristics. However, these cases showed positive immunoreactivity to calponin, CD10, p63, and vimentin, which helped determine the correct diagnosis of myoepithelial carcinoma. Immunoreactivity for C-kit was negligible, seen in only 6% of cases (3 of 50) and was focal. Only 6 of 50 carcinomas expressed immunoreactivity for Ki-67, with the Ki-67 labeling index ranging from 4% to 10%. Four of these 6 cases were high-grade tumors, and 2 were intermediate-grade carcinomas.

ULTRASTRUCTURAL FEATURES

Ultrastructural examination was performed in 2 cases. Pure spindle-cell morphologic characteristics were noted in one case, and mixed epithelioid and plasmacytoid morphologic characteristics in the other. Both cases showed presence of reduplicated basal lamina, numerous intracytoplasmic intermediate filaments, fine actin filaments, and cell junctions (Figure 4).

TREATMENT

Following the initial diagnosis, 44 of the 51 patients in this study with myoepithelial carcinomas underwent surgical excision. The remaining 7 patients, who had visited our institution only for a second opinion, were lost to follow-up. Postoperative radiotherapy was given to 12 patients, while none received chemotherapy. Excision of the primary tumor along with cervical lymph node dissection was performed in 17 patients.

FOLLOW-UP

Eighteen patients who underwent excision of the primary tumor developed recurrence; the time to recurrence ranged from 2 months to 11 years. Of these 18 tumors, 11 were intermediate-grade carcinomas, 6 were low-grade, and 1 was a high-grade carcinoma. Multiple recurrences were seen in 5 patients.
Recurrence was common among tumors arising in the buccal mucosa (4 of 7, 57%), parotid gland (7 of 15, 47%), and nasal cavity (2 of 5, 40%). A high incidence of recurrence was noted with the cell types stellate (5 of 8, 63%), clear (2 of 3, 67%), and spindle (3 of 6, 50%). Among the 12 carcinomas associated with a preexisting benign tumor, 5 (3 myoepitheliomas and 2 pleomorphic adenomas) showed recurrence. Recurrence was also noted in 3 of 4 patients who showed perineural invasion. Eleven of 38 tumors in the size range of 1 to 5 cm showed recurrence (29%), while 6 of 10 tumors in the size range of 6 to 10 cm showed recurrence (60%). Therefore, it can be stated that the size of the tumor is associated with recurrence. Recurrence was also seen in 83% of tumors that showed bone invasion (5 of 6).

Cervical lymph node metastasis was observed in 7 of 17 cases in which lymph node dissection was performed (41%) (Figure 5). Of these 7, 4 were associated with recurrence, and 3 were associated with distant metastasis. The metastatic sites were thigh musculature, brain, and vertebral bones. One of these 7 cases was a high-grade carcinoma, 4 were intermediate-grade, and 2 were low-grade carcinomas. Five of these 7 cases (71%) demonstrated either pure spindle-cell morphologic characteristics (2 cases) or spindle cell mixed with other cell types (3 cases) both in primary and metastatic tumors. The remaining 2 cases showed pure epithelioid cell morphologic characteristics. Tumors with greater than 50% necrosis, positive margins, high mitotic count (>4 per 10 hpf), a Ki-67 labeling index of 4% to 10%, nuclear atypia, and spindle cell morphologic characteristics showed a high frequency of metastasis.

Adequate clinical follow-up (minimum 24 months) was available for 18 patients who were primarily treated in the hospital. Of these 18 cases, 12 showed recurrence, 3 showed lymph node metastasis, and 2 showed distant metastasis. None of these 18 patients died of disease. Fifteen patients were without any evidence of disease after a mean of 63.73 months (range, 24-120 months). The remaining 3 patients were alive with disease at 34, 98, and 120 months, respectively, although 2 of them had recurrence, and 1 had brain and lymph node metastasis. Using the Kaplan-Meier survival curve, we found the mean disease-free survival time to be 31.9 months (Figure 6).

The effect of various tumor parameters (size, site, growth pattern and architecture, pattern of infiltration, cell type, stroma, nuclear atypia, mitosis, necrosis, perineural invasion, margin or bone involvement, and evidence of preexisting benign tumor) on the prognosis was studied statistically using the $\chi^2$ test. None of the factors showed a significant correlation.
As noted by Savera and Zarbo, myoepithelial cells were first described by Zimmerman in 1898. Myoepithelial cells are believed to be of ectodermal origin. They envelop the glandular, acinar, and ductal elements of various organs, especially breast and salivary glands. In salivary glands, the myoepithelial cells that surround the intercalated ducts are spindled in contrast to the large stellate ones that envelop the acini. 

Myoepithelial salivary gland neoplasm was first recognized in 1943 but recent reviews by contemporary investigators—including Barnes et al, Scuibba and Bran- non, Dardick and van Nostrand,10 Dardick et al, and Dardick—have renewed awareness of this form of salivary gland neoplasia, defining many of its cytomorphologic, ultrastructural, and immunophenotypic characteristics. Myoepithelial salivary gland tumors constitute 1% of all salivary gland tumors. The relatively recently recognized and rarely encountered malignant myoepithelial tumor (ie, myoepithelial carcinoma) was first described by Stromeyer et al11 in 1975 and has been included in the World Health Organization classification of salivary gland tumors since 1991. A review of the world literature in 2000 disclosed approximately 75 cases of myoepithelial carcinoma. Some of these carcinomas originated de novo whereas others were classified myoepithelial carcinoma ex pleomorphic adenoma or as arising in benign myoepitheloma.

The currently accepted diagnostic criteria for myoepithelial carcinoma are exclusive myoepithelial differentiation (morphologic and IHC) and clear-cut tumor infiltration into adjacent salivary gland or other tissues. Assessment of invasion is usually difficult in tumors arising within minor salivary glands, and this requires adequate sampling of the tumor-host tissue interface, which may not be included within a biopsy specimen. In the present study, all of the tumors displayed tumor infiltration, the extent varying from case to case, but general features of malignancy such as marked cytologic atypia with necrosis were seen only in a proportion of neoplasms. Recently, Nagao et al suggested that a high cell proliferative activity (ie, 7 mitosis instances per 10 hpf or a Ki-67 labeling index >10%) may indicate malignancy. In the present series, tumors with a Ki-67 labeling index of 4% to 10% showed a high frequency of metastasis.

Most studies have found that 75% of myoepithelial carcinomas arise in the major salivary glands and that about half arise in precursor lesions. However, the results in the present study were quite different: minor salivary gland involvement (71%, n = 36) was more common than major salivary gland involvement (29%, n = 15). Also precursor benign lesions were seen only in 24% of cases (n = 12). Sometimes, it is very difficult to identify the pre-existing benign component within the tumor, especially when the tumor is low grade. Myoepithelial carcinomas arising within a preexisting benign tumor should be suspected if there is long history of benign parotid tumor with history of rapid growth and/or multiple recurrences in a preexisting pleomorphic adenoma with or without lymph node metastasis.

Differentiation into different morphologic cell types is a unique feature of myoepithelial cells, and this property has been well described in the present study. In addition, we observed that in certain cases, the tumor cells were ovoid to short spindly with centrally placed nuclei, a moderate amount of cytoplasm, and indistinct cell borders. The cells were arranged in a diffuse, sheetlike pattern. This cell type could be called neither epithelioid nor spindle cell and was labeled as stellate type. This distinction is essential because 5 of 8 cases that showed stellate morphologic characteristics in present study showed recurrence (63%), thus suggesting an association between a high rate of recurrence and this morphologic characteristic. Vacuolated or signet ring cell morphologic characteristics were not observed in this series.

In the present series, a diffuse sheetlike growth pattern was the most common, followed by a multinodular pattern. In contrast, Savera at found that the multinodular pattern was the most common growth pattern, thought to be due to high levels of diverse proteinase inhibitors. Normal myoepithelium is capable of synthesizing basement membrane components and extracellular matrix, and this property is augmented by neoplastic myoepithelium resulting in the formation of chondroitin sulfate proteoglycan (bluish-gray myxochondroid matrix), type IV collagen, laminin, fibronectin, and types I and II collagen (eosinophilic hyaline matrix). However, matrix production with formation of stroma was not seen in tumors showing pure spindle-cell morphologic characteristics.

The IHC studies in the present study fully confirmed myoepithelial differentiation, with 80% of tumors overall expressing vimentin, S-100 protein, and calponin together. Vimentin and S-100 protein are usually not present in normal myoepithelial cells and are very sensitive (but not specific) markers of neoplastic myoepithelium. Neoplastic myoepithelium has also been known to lose or modify its smooth-muscle phenotype, and hence IHC studies to demonstrate myogenic differentiation might not always be helpful. In recent years, calponin has been proved to be a specific and fairly sensitive marker of myoepithelial differentiation in salivary gland tumors. In the present cases with unusual morphologic characteristics (8 of 50) and in which findings were negative for S-100 protein, SMA, and CK, positive reactions to antibodies like calponin, CD10, p63, and vimentin proved helpful in diagnosing myoepithelial carcinoma correctly. Although noted in only 28% of tumors in present study (n = 14), p63 has been reported to be a useful marker of myoepithelial cells in salivary gland neoplasms. But p63 is also expressed by squamous cell carcinoma and mucoepidermoid carcinoma. In addition, more than 1 CK antibody (eg, AE-1 and -3, CK-5 and -6, 34-BE12) should be used for testing because neoplastic myoepithelium usually tests negative for CK-7 and EMA. Therefore, a high index of suspicion and confirmation via a number of IHC markers are necessary for diagnosis of myoepithelial carcinoma, especially in settings of pure spindle-cell or plasmacytoid morphologic characteristics with rhabdoid differentiation. A low incidence of C-kit reactivity, as observed in our study, was also noted by Stelow et al and Jeng et al.
The differential diagnosis of myoepithelial carcinoma includes a wide range of neoplasms, depending on the predominant cell type. It is sometimes difficult to differentiate myoepithelial carcinoma showing epithelioid morphologic characteristics from other salivary gland neoplasms showing myoepithelial differentiation, especially adenoid cystic carcinoma, polymorphous low-grade carcinoma, and others. Demonstration of luminal differentiation by CEA and EMA immunostaining favors diagnosis of adenoid cystic carcinoma.6 In tumors with clear-cell morphologic characteristics, the differential diagnosis includes hyalinizing clear-cell carcinoma, epimyoepithelial carcinoma, and metastatic renal cell carcinoma.22 Melanoma, high-grade lymphoma, or plasmacytoma must be ruled out when the tumor shows plasmacytoid differentiation. With spindle-cell morphologic characteristics, the most common differentials are sarcomatoid squamous carcinoma, spindle-cell melanoma, and schwannoma. To help distinguish between them, the diagnostician should use specific morphologic criteria like melanin pigment, prominent cosinophilic nuclei (melanoma), a biphasic pattern with luminal differentiation (adenoid cystic carcinoma and epimyoepithelial carcinoma), Antoni A and Antoni B areas (schwannoma), and an in situ squamous component (sarcomatoid squamous carcinoma) in combination with appropriate IHC analyses.6 At times, diagnosis of myoepithelial carcinoma remains challenging, with many factors contributing to the challenge. These include a preexisting benign lesion in the patient’s history and cytomorphologic heterogeneity.22,26

To our knowledge, no substantial evidence has been reported to predict clinical behavior or outcome of tumors on the basis of histologic features. In the present study, a 3-tier grading system was used to analyze the difference in the clinical behavior in different grades. However, our results showed a wide distribution of grades in relation to clinical behavior. Recurrence was noted in 18 cases, of which only 1 was a high-grade tumor, while 11 were intermediate grade, and 6 were low grade. Other reported cases with low-grade features have resulted in widespread metastasis and patient death,20 while some histologically aggressive variants were associated with long-term survival.5,13 This raises an important question: Should pathologists attempt to grade myoepithelial carcinomas? According to Yu et al,30 myoepithelial carcinomas of the salivary gland should be classified as high-grade malignant neoplasms with a poor prognosis.30 However, DiPalma and Guzzo12 considered myoepithelial carcinoma to be a low-grade cancer when it arose in a pleomorphic adenoma and high grade when it was de novo. Such a difference was not reflected in our study. In the present study, the results did not entirely support the 3-tier grading system, and hence combining intermediate-grade with high-grade tumors and using a 2-tier system (low grade and high grade) would be reasonable.

Furthermore, in the present study, a high incidence of recurrence was noted with stellate, clear, and spindle-cell types; large tumor size; and perineural or bone invasion. Similarly, a high incidence of metastasis was noted with the presence of positive margins, large areas of necrosis, high mitotic count (>4 per 10 hpf), Ki-67 labeling index of 4% to 10%, nuclear atypia, and plasmacytoid cell morphologic characteristics. However, Savera et al6 in their series of 25 cases, found only a weak statistical correlation for outcome with cytologic atypia (high grade), but other parameters (tumor size, site, cell type, mitotic rate, presence of a benign tumor, necrosis, and perineural and vascular invasion) showed no relationship. Therefore, we believe that it would be best to list the various histologic features to grade the tumor as low or high while at the same time noting that histologic grade is still a far from proven guide to clinical behavior. Table 4 provides data for comparison between the present study and 2 other major studies.

The treatment of myoepithelial carcinoma has been mainly surgical, including wide excision with free margins, with or without nodal dissection.6,13 The role of chemotherapy and radiotherapy has not yet been established.

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Table 4. Literature Findings on Studies of Myoepithelial Carcinoma Compared With Those of the Present Studya

<table>
<thead>
<tr>
<th>Study</th>
<th>Total Cases</th>
<th>Patient Age Range, y</th>
<th>Tumor Site</th>
<th>CC, NA N PNI</th>
<th>Assoc BT</th>
<th>Prognosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Present study</td>
<td>51</td>
<td>14-70</td>
<td>Par, 15 Pal, 15</td>
<td>E, 15</td>
<td>21</td>
<td>18</td>
</tr>
<tr>
<td>Nagao et al13</td>
<td>10</td>
<td>48-81</td>
<td>Par, 7 Pal, 7</td>
<td>Mixed, 6 E+P; 3 E+S; 3</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Savera et al6</td>
<td>25</td>
<td>24-77</td>
<td>Par, 15 Pal, 2</td>
<td>E, 11</td>
<td>10</td>
<td>15</td>
</tr>
</tbody>
</table>

Abbreviations: Assoc BT, associated benign tumor; CC, common-cell type; DOD, died of disease; E, epithelioid; FU, follow-up; LFU, lost to FU; M, myoepithelioma; METS, metastasis (nodal + distant); N, necrosis; NA, nuclear atypia; P, plasmacytoid; PA, pleomorphic adenoma; Pal, palate; Par, parotid; PNI, perineural invasion; REC, recurrence; S, spindle.

aUnless otherwise indicated, all numerical data indicate number of cases.
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Author Contributions: Both authors had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. Study concept and design: Kane and Bagwan. Acquisition of data: Bagwan. Analysis and interpretation of data: Kane and Bagwan. Drafting of the manuscript: Bagwan. Critical revision of the manuscript for important intellectual content: Kane and Bagwan. Statistical analysis: Bagwan. Administrative, technical, and material support: Bagwan. Study supervision: Kane.

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