INTRODUCTION

Almost 2000 indexed articles in the relevant literature of the last 10 years deal with the exciting topic of the diagnosis, behavior (recurrence, malignant transformation), and treatment of pleomorphic adenoma (PA) (Available at: 94420180https://pubmed.ncbi.nlm.nih.gov/?term=%28%22pleomorphic+adenoma%22%29+AND+%28%222011%2F01%22%5BDatetime+Entry%5D+%3A+%223000%22%5BDatetime+Entry%5D%29). Undoubtedly, the mostly incurable multifocal recurrence in parotideal cases, the malignant transformation with a critical prognosis, and the risk of treatment-related iatrogenic damage to the facial nerve make this entity challenging from a clinical point of view.

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ORIGINAL ARTICLE

Histopathological comparison of pleomorphic adenomas of the parotid and submandibular gland

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Abstract

Objective: The aim of this study was to investigate and compare the histopathological findings in pleomorphic adenomas (PA) of the parotid and submandibular gland with emphasis on the histological subtype and capsular characteristics.

Materials and Methods: The histopathological specimens of all patients with PAs of the parotid and submandibular gland between 2000 and 2020 were re-examined by an experienced head and neck pathologist. Patients without representative slides allowing evaluation of the whole periphery of the PA were excluded from our study sample.

Results: Nine hundred and thirty-four patients formed our study sample (327 men, 607 women, male-to-female ratio: 0.53:1). Eight hundred and forty-four cases had a PA in the parotid gland and the remaining 90 in the submandibular gland. Our comparative analysis showed that submandibular PAs are characterized by the consistent presence of an intact anatomical capsule, infrequent occurrence of pseudopodia and satellite nodules, and a low proportion of the high-risk myxoid subtype.

Conclusion: Our study highlights significant differences between PAs of the parotid and submandibular glands in their histopathological characteristics. Their differences likely underlie the favorable surgical outcome observed in PAs of the submandibular glands and may explain the propensity of PAs of the parotid glands for local recurrences.

Keywords

capsule, parotid gland, pleomorphic adenoma, satellite nodules, submandibular gland
A pseudopodium represents a "re-encapsulated" secondary nodule of the tumor tissue by an anatomically intact fibrous capsule. Lack of certainty on this issue complicates the therapeutic management of this disease.

Furthermore, it is known that parotideal PAs show a significant long-term recurrence rate of up to 7% (if followed up for 20 years; Valstar et al., 2020), whereas their submandibular counterparts recur only extremely rarely (Mantsopoulos et al., 2017). In trying to explain this remarkably different biological behavior of the same lesion in different organs, the aim of this study was to compare the cases with PAs of the parotid and the submandibular glands in terms of epidemiological factors (age, gender) as well as the incidence of the various histopathological parameters (histological subtype, intactness of the capsule, surgery-induced capsular defects, pseudopodia (PS), satellite nodules [SN]).

2 | MATERIALS AND METHODS

This study was performed at an academic tertiary referral center specialized in salivary gland diseases (Department of Otorhinolaryngology, Head and Neck Surgery, University of Erlangen–Nuremberg, Erlangen, Germany). For this study, an experienced head and neck pathologist (A.A.) criticized re-evaluated the histological slides of the pathological specimens of all patients who underwent primary submandibulectomy for PAs of the submandibular gland or parotidectomy for PAs of the parotid gland between 2000 and 2020. Based on tumor size, the number of histological slides per tumor varied from one to more than 10. The 38 cases with PAs in which the whole periphery of the tumor was not histologically ascertainable on at least two slides, based on the tumor size, were excluded from the study sample.

Concerning the histopathological analysis, PAs were divided into 3 histological subtypes on the basis of the stroma cell proportion, according to the classification of Seifert et al., (1976): the classic "mixed" subtype with a stroma content of 30%-50%, the stroma-rich (myxoid) subtype with a stroma content of more than 50%, and the cellular subtype with a stroma content of 30% or less. Furthermore, we paid special attention to the specific characteristics of the capsule likely to be associated with possible recurrence, such as the presence of a complete and intact tumor capsule, surgically induced capsular defects, PS, and SN. For comparison with the relevant literature, we used the nomenclature proposed by Zbaren & Stauffer, (2007): a complete capsule assumed complete encapsulation of the tumor tissue by an anatomically intact fibrous capsule. A pseudopodium represents a "re-encapsulated" secondary nodule separated only by a layer of capsular fibrous tissue from the main tumor mass, but localized within the main tumor capsule. Based on the plan of sectioning, PS may appear either as a mushroom-like capsular protrusion or as a distinct small nodule closely associated with the main tumor and separated from it only by the fibrous capsule, but no normal fatty or salivary tissue is seen between the two nodules. Finally, SN were distinct tumor nodules near the main tumor but outside the main tumor capsule, separated from it by salivary or fatty tissue without any connection to the main tumor.

Statistical analysis was performed using the T test for the epidemiological analysis and the chi-square test for the comparison of histological parameters with 95% confidence intervals (CIs). The software SPSS version 21 for Windows (SPSS, Inc.) was used for the analysis. A p-value of <.05 was considered statistically significant. The Institutional Review Board (IRB) of the University Hospital of Erlangen approved this study.

3 | RESULTS

Nine hundred and thirty-four patients formed our study sample (327 men, 607 women, male-to-female ratio: 0.53:1). Eight hundred and forty-four cases had a PA in the parotid gland, and the remaining 90 had a PA in the submandibular gland. The mean age was 48.6 (12–89 years). No statistically significant differences were detected in gender (p = .906) or age (p = .282) between patients with PAs of the parotid and submandibular glands. Table 1 shows the results of the statistical analysis of the histopathological parameters in our study cases. Comparative analysis showed that PAs of the submandibular gland are characterized by the consistent presence of an anatomically intact capsule, a significantly lower occurrence of PS, a tendency toward a lower incidence of satellite tumors, and a comparatively limited incidence of the fragile myxoid subtype.

4 | DISCUSSION

A significant number of literature reports try to explain the exact pathogenesis of PA recurrence based on the interaction between several tumor- and treatment-related factors. Although submandibulectomy tends to lead quite frequently to a broad exposure of the capsule of a submandibular PA, it is common experience that submandibular lesions recur extremely rarely (Mantsopoulos et al., 2017). On the contrary, although a vast amount of literature reports are dedicated to the need for preserving a cuff of healthy parotid tissue around a parotid PA (Mantsopoulos et al., 2021), long-term recurrence rates of these lesions are by no means negligible (Valstar et al., 2020). The lack of adequate treatment justifies the characterization of the most common multifocal recurrence of a parotid PA as a so-called "benign tragedy" (Poorten, 2014). This apparent difference in the biological behavior between parotideal and submandibular PA s prompted us to investigate the histological differences ("tumor-related factors") between these lesions in a large
single-center cohort in order to obtain information that could help to enlighten the recurrence mechanism.

PA is one of the most common neoplasms of the parotid gland (Mantsopoulos & Iro, 2020). The prefix “pleomorphic” reflects the architectural diversity of these lesions due to highly variable combinations of epithelial and myoepithelial elements, intermingled with a similarly variable mucoid, myxoid, or chondroid mesenchymal component. A review of the relevant literature shows a significant diversity in the incidence of several subtypes (myxoid, mixed, hypercellular; Seifert et al., 1976) in parotideal PAs, probably due to differences in the classification systems (Dulguerov et al., 2017). Re-examination of our parotid specimens showed that almost half of them belonged to the myxoid subtype (47.7%), with a homogenous distribution of the remaining lesions in the other categories.

**TABLE 1** Analysis of the histopathological parameters in pleomorphic adenomas of the parotid and submandibular gland in our study (bold values: statistically significant for \( p < 0.05 \), bold italic values: tendency to statistical significance)

| Histopathological features | Parotid gland (%) | Submandibular gland (%) | Total (%) | \( p \) |
|---------------------------|-------------------|-------------------------|-----------|-------|
| **Histological subtype**  |                   |                         |           |       |
| Hypercellular subtype (stroma content < 30%) | 206 (24.4) | 20 (22.2) | 226 (24.2) | 0.000 |
| Mixed subtype (stroma content 30%-50%) | 235 (27.8) | 45 (50) | 280 (30) |       |
| Stroma-rich (myxoid) subtype (stroma content > 50%) | 403 (47.8) | 25 (27.8) | 428 (45.8) |       |
| **Intact capsule** |                   |                         |           |       |
| No capsule at all         | 36 (4.3)          | 0 (0)                   | 36 (3.9)  | 0.000 |
| Partial encapsulation     | 118 (14)          | 0 (0)                   | 118 (12.6) |       |
| Intact capsule            | 690 (81.7)        | 90 (100)                | 780 (83.5) |       |
| **Pseudopodia**           |                   |                         |           |       |
| Presence of pseudopodia   | 367 (43.5)        | 17 (18.9)               | 384 (41.1) | 0.000 |
| No pseudopodia            | 477 (56.6)        | 73 (81.1)               | 550 (58.9) |       |
| **Satellite nodules**     |                   |                         |           |       |
| Presence of satellite nodules | 67 (7.9) | 3 (3.3) | 70 (7.5) | 0.077 |
| Absence of satellite nodules | 777 (92.1) | 87 (96.7) | 864 (92.5) |       |
| **Surgical defect of the capsule** |           |                         |           |       |
| Surgically induced defect of the capsule | 37 (4.4) | 2 (2.2) | 39 (4.2) | 0.255 |
| No surgically induced defect of capsule | 807 (95.6) | 88 (97.8) | 895 (95.8) |       |

Bold values are statistically significant for \( p < 0.05 \), bold italic values are tendency to statistical significance.

**FIGURE 1** (a, b) Pleomorphic adenomas (PA) of the parotid gland with pseudopodia (PS) and satellite nodules (SN)
(hypercellular in 24.4% and “mixed” type in 27.8%). Contrary to this, the majority of submandibular cases manifested the mixed subtype (50%), which showed a statistically significant difference from their parotid counterparts ($p = .000$). It seems that less stroma content in the submandibular cases warrants more stability and less fragility in comparison with the parotid lesions.

PA is the sole benign lesion in the parotid gland with a frequently instable capsule with herniation of tumor material in the surrounding healthy glandular parenchyma (Figure 1). The capsule is a thin layer of fibrous connective tissue with a variable morphology, in terms of thickness and integrity. Analyzing a series of 100 cases, Stennert et al. found that the capsule of PAs had a maximal thickness of 250 μm (Stennert et al., 2001). In our series of 844 parotideal PAs, only three of our 844 cases (0.3%) reached focally a capsule thickness of 500 μm. Concerning intactness of the capsule, the potential correlation of a capsule defect with the recurrence of PAs has already been described for almost 60 years (Patey & Thackray, 1958). Several literature reports point to the fact that a significant amount of PAs could exhibit at least partial defects (Dulguerov et al., 2017; Stennert et al., 2001). Our series was no exception to this common histopathological experience: 13.9% of our parotid cases showed anatomical defects of the capsule, whereas another 4.2% of the same group showed no capsule elements at any site on the periphery of the PA, as is typically seen in PAs of the minor salivary glands. In total, almost one-fifth (18.1%) of our parotid cases exhibited partial or complete absence of the capsule. Interestingly, submandibular PAs had an intact capsule in all of our 90 study cases without exception ($p = .000$).

Another histopathological feature with clinical relevance refers to the presence of surgically induced defects of the capsule (Mantsopoulos & Iro, 2021). In the parotid gland cases, close dissection in the vicinity of critical structures (facial nerve) or in the tumor–nerve interface can lead to an accidental rupture of the adenoma capsule. In submandibular cases, common surgical experience shows that adenomas usually lie in the peripheral zone of the gland (Figures 2 and 3). The need for avoiding injury to the marginal mandibular branch of the facial nerve on the cranial aspect and the lingual nerve on the medial aspect of the gland dictates a subcapsular dissection of the gland outside its bed (in the course of submandibulectomy; Mantsopoulos et al., 2017, 2018), which exposes a PA lying in the gland periphery with a high risk of rupture. Such microscopic defects could be seen in 4.2% of our parotid and 2.2% of our submandibular cases, without a statistically significant difference between these study groups ($p = .255$). This points to the fact that the localization of a lesion in the gland is the most important risk factor for a surgically induced defect of its capsule.

In the 1950s, Patey and Thackray already recognized the value of PS in the pathomechanism of the recurrence of Pas (Patey & Thackray, 1958). The incidence of PS in parotideal PAs varies between 28 (Stennert et al., 2001) and 54% (Park et al., 2012) in the relevant literature. This remarkable diversity of percentages points to the value of accuracy in histopathological examination. In our sample, 43.5% of the cases in the parotid gland showed this feature. Interestingly, PS presented in only 18.8% of the submandibular PAs, showing a statistical significance in comparison with parotid gland lesions. The incidence of satellite tumors in the vicinity of PAs varies by 3% (Orita et al., 2010), 13% (Zbaren & Stauffer, 2007), and 28% (Stennert et al., 2001) in the relevant literature. The great variability of scores reflects the histopathological pitfall of confusing the tip of the pseudopodium with a satellite nodule under microscopic view. Thus, one cannot rule out that the SN in several of these cases may have actually represented PS projecting into the surrounding parotid tissue, giving the impression of being a satellite nodule. This points to the need to examine several slides of the same lesion. In our series, 7.9% showed this feature with a varying number of lesions and distances from the main tumor. Comparative analysis of parotideal and submandibular cases showed a lower incidence of this feature in the submandibular cases (3.3%) with a tendency toward significance (0.07).

In the largest single-center comparative histopathological analysis of PAs of the major salivary glands, we found significant differences in terms of histological subtypes and capsular characteristics between parotid and submandibular gland lesions. PAs of the submandibular gland are characterized by the consistent presence of an anatomically intact capsule, a remarkably low occurrence of PS and satellite tumors, as well as a comparatively limited incidence of the fragile myxoid subtype (Figure 1). These histological characteristics have an apparent surgical relevance: an anatomically intact capsule ensures a firm encapsulation and sufficient margination of the lesion from the surrounding tissue. A lack of PS ensures a smooth, even surface, whereas the limited incidence of the fragile type makes these adenomas more stable and improves their tolerance of surgical manipulations. Common surgical experience shows that the majority of submandibular PAs...
lie in the peripheral part of the gland (apparently mostly due to relatively small size of the gland, Figure 3) and their capsule is incorporated in (or replaces) the gland capsule (Figure 2). In this most frequent scenario, the aforementioned histological characteristics seem to act protectively and prevent opening of a peripheral tumor during surgical dissection along the capsule of the submandibular gland (Figure 3).

Provided that the surgical principle “do not see the capsule” is strictly adhered to, it seems that rather the localization and size of the PA determine the extent of capsule exposure: a most frequently eccentrically localized submandibular PA has the same risk of exposure of its capsule as a large PA of the parotid with broad contact to the facial nerve. Leaving surgery-related factors aside, it seems that the rather stable and comparatively beneficial as well as more predictable histological profile of submandibular cases could explain their generally accepted favorable prognosis.

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CONFLICT OF INTEREST
None.

AUTHOR CONTRIBUTIONS
Konstantinos Mantsopoulos: Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Project administration; Software; Validation; Visualization; Writing – original draft; Writing – review and editing. Matti Sievert: Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Project administration; Resources; Supervision; Validation; Visualization; Writing – original draft; Writing – review and editing. Abbas Agaimy: Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Project administration; Resources; Supervision; Validation; Visualization; Writing – original draft; Writing – review and editing. Heinrich Iro: Conceptualization; Project administration; Resources; Software; Supervision; Writing – review and editing.

REFERENCES
Dulguerov, P., Todic, J., Pusztaszeri, M., & Alotaibi, N. H. (2017). Why do parotid pleomorphic adenomas recur? A systematic review of pathological and surgical variables. *Front Surg*, 4, 26. https://doi.org/10.3389/fsurg.2017.00026
Mantsopoulos, K., Goncalves, M., Koch, M., Iro, H., & Agaimy, A. (2018). Submandibular gland pleomorphic adenoma: Histopathological capsular characteristics and correlation with the surgical outcome. *Annals of Diagnostic Pathology*, 34, 166–169. https://doi.org/10.1016/j.ioandopath.2018.04.002
Mantsopoulos, K., Goncalves, M., Schapher, M., & Iro, H. (2017). Long-term oncologic and functional outcome in pleomorphic adenomas of the submandibular gland. *Laryngoscope Investig Otolaryngol*, 2, 369–372. https://doi.org/10.1002/lio2.113
Mantsopoulos, K., Iro, A. K., Sievert, M., Müller, S. K., Agaimy, A., Koch, M., & Iro, H (2021). Refinement of the surgical indication and increasing expertise are associated with a better quality of pathology specimens in pleomorphic adenomas. *Acta Oto-Laryngologica*, 1–5. https://doi.org/10.1080/00016489.2021.1871947
Mantsopoulos, K., & Iro, H. (2020). Pleomorphic adenoma compared with cystadenolymphoma of the parotid gland: Which is more common? *British Journal of Oral and Maxillofacial Surgery*, 58, 361–363. https://doi.org/10.1002/lio.20201
Mantsopoulos, K., & Iro, H. (2021). Tumour spillage of the pleomorphic adenoma of the parotid gland: A proposal for intraoperative measures. *Oral Oncology*, 112, 104986. https://doi.org/10.1016/j.oraloncology.2020.104986
Orita, Y., Hamaya, K., Miki, K., Sugaya, A., Hirai, M., Nakai, K., Nose, S., & Yoshino, T. (2010). Satellite tumors surrounding primary pleomorphic adenomas of the parotid gland. *European Archives of Oto-Rhino-Laryngology*, 267, 801–806. https://doi.org/10.1007/s00415-009-1149-7
Park, G. C., Cho, K. J., Kang, J., Roh, J. L., Choi, S. H., Kim, S. Y., & Nam, S. Y. (2012). Relationship between histopathology of pleomorphic adenoma in the parotid gland and recurrence after superficial parotidectomy. *Journal of Surgical Oncology*, 106, 942–946. https://doi.org/10.1002/jso.23202
Patey, D. H., & Thackray, A. C. (1958). The treatment of parotid tumours in the light of a pathological study of parotidectomy material. *British Journal of Surgery*, 45, 477–487. https://doi.org/10.1002/bjs.18004519314

Poorten, V. V. (2014). Recurrent-residual pleomorphic adenoma: the benign tragedy IFHNOS 2014.

Seifert, G., Langrock, I., & DONath, K. (1976). A pathological classification of pleomorphic adenoma of the salivary glands (author’s transl). *HNO*, 24, 415–426.

Stennert, E., Guntinas-Lichius, O., Klussmann, J. P., & Arnold, G. (2001). Histopathology of pleomorphic adenoma in the parotid gland: A prospective unselected series of 100 cases. *Laryngoscope*, 111, 2195–2200. https://doi.org/10.1097/00005537-20011200-00024

Valstar, M. H., Andreasen, S., Bhairosing, P. A., & McGurk, M. (2020). Natural history of recurrent pleomorphic adenoma: Implications on management. *Head and Neck*, 42, 2058–2066. https://doi.org/10.1002/hed.26137

Zbaren, P., & Stauffer, E. (2007). Pleomorphic adenoma of the parotid gland: Histopathologic analysis of the capsular characteristics of 218 tumors. *Head and Neck*, 29, 751-757. https://doi.org/10.1002/hed.20569

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