Int J Clin Exp Pathol 2020;13(8):2050-2057
www.ijcep.com /ISSN:1936-2625/IJCEP0115409

Original Article

The clinicopathological features of parotid lymphoma

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Received June 1, 2020; Accepted July 20, 2020; Epub August 1, 2020; Published August 15, 2020

Abstract: Lymphoma of the parotid gland is a rare malignant tumor, and cohort studies on the survival rates of affected patients are sparse. This study aimed to retrospectively evaluate the clinicopathological characteristics of patients diagnosed with non-Hodgkin lymphoma of the parotid gland. This study included 31 patients diagnosed with lymphoma of the parotid gland. Data on the pathological subtypes, the WHO classifications of the hematopoietic and lymphoid tissues, and the Ann Arbor staging, treatment modalities, and survival times were collected and analyzed. Among the 31 patients, there were 18 cases of extranodal marginal zone B-cell lymphoma of the mucosa-associated lymphoid tissue (MALT), 7 cases of diffuse large B-cell lymphoma (DLBCL), and 6 cases of follicular lymphoma (FL). The tumors were most-commonly located in the superficial lobe of the parotid gland (28/31), and three cases involved the deep lobe of the parotid gland (3/31). The overall median survival from the diagnosis of lymphoma was estimated to be 62 months, with 3-year and 5-year survival rates of 83.9% and 77.4%, respectively. A univariate analysis demonstrated statistically significant differences in accelerated tumor growth ($P<0.001$) and the presence of tumor capsules ($P<0.001$). A multivariate analysis demonstrated statistically significant differences in the accelerated tumor growth ($P=0.029$). MALT lymphoma was the most common subtype of primary parotid lymphoma. The prognosis is better than it is with other malignant parotid tumors. The presence of accelerated tumor growth was significantly correlated with overall survival time.

Keywords: Lymphoma, parotid gland, clinical pathology, prognosis

Introduction

The occurrence of primary salivary gland lymphoma is rare, accounting for about 2% of all salivary gland tumors, 75% of which occur in the parotid gland [1]. The oral maxillofacial region and the head and neck region are the second most common malignant lymphoma sites [2], and other sites that may be affected are the ocular adnexal and the thyroid [3].

In the early stages of parotid lymphoma, patients are generally asymptomatic, but when patients develop facial symptoms including facial pain, facial paralysis, and growth acceleration they tend to visit physicians [4]. When patients' facial symptoms are initially observed, they generally are first referred to the department of stomatology rather than the department of hematology, resulting in most parotid lymphoma being initially treated as any other parotid malignancy [5]. *The WHO Classification of Tumours of Haematopoietic and Lymphoid Tissue* staging system is usually used for tumor staging before surgery [6]. It is difficult to confirm the presence of parotid lymphoma before surgery, so many cases are diagnosed as lymphoma after tumor resections [5]. The definitive diagnosis is made through a pathological examination of the surgical specimens. The staging standard used by most hematologists in extranodal lymphoma cases is the Ann Arbor staging system [7, 8]. We present 31 cases of parotid lymphoma whose diagnosis was confirmed using immunohistochemistry. The histological classification was based on the World Health Organization classification standard of hematopoiesis and lymphoid tissue tumor diseases [9], which is based on the Ann Arbor staging system [8]. Extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue (MALT) and follicular lymphoma (FL) are the most common histological classifications of parotid lymphoma [10] which

has many distinctive pathological features and a better prognosis than other malignant tumors. The absence of a tumor capsule indicates a poor prognosis. This study determined the association between staging and clinicopathology. In addition, we determined the association between the survival times of patients who underwent surgery and those who did not.

Materials and methods

Case selection

Our study on parotid lymphoma was carried out between January 2010 and January 2015. The basic information recorded from each patient which included gender, age, local symptoms, the pathological subtype of NHL, whether or not it was accompanied by an autoimmune disease condition, the B symptoms, the degree of abnormal serum lactate dehydrogenase (LDH) elevation, the clinical-stage, treatment modalities, chemotherapy regimens, International Prognostic Index (IPI) scores, and the 3-year and 5-year survival rates. The standard approach used to assess the prognoses of those patients with lymphoma was IPI [11], and the score was based on age, extranodal sites, physical condition, clinical staging, and serum lactate dehydrogenase levels at four levels: 0-1 points for low risk, 2 points for low-intermediate risk, 3 points for high-intermediate risk, and 4-5 points for high risk. The overall survival (OS) was calculated based on the date of diagnosis and the date of death or the loss of follow-up, or the last follow-up for any cause (months). We included 31 patients diagnosed with parotid lymphoma as confirmed by immunohistochemistry based on histopathological investigations by two senior pathology experts and paraffin sections with an immunohistochemical analysis.

Immunohistochemical analysis and evaluation of staining

The immunohistochemical analysis was performed using Elivision™ Plus detection kits (LabVision, USA), and we examined the CD20, CD10, Pax-5, Bcl-2, Bcl-6, MUM-1, CD79a, cyclinD1, and Ki-67 expressions. The scores were based on the extent and intensity of the staining, >3 was positive, ≤3 was negative. The classification of the positive staining was as follows: 1, ≤10%; 2, 11%-50%; 3, 51%-75%; and 4, >75%. The staining intensity was divided into four grades as follows: 0, none; 1, weak; 2, moderate; and 3, strong. The staining results were interpreted by two pathologists who were blinded to the clinical data and were determined by semi-quantitative points. When there was a disagreement among the observers, a consensus was reached through a re-examination of the sections.

Statistical analysis

To determine the median, 3-year, and 5-year OS, Kaplan-Meier curves were generated, and log-rank tests were employed for the comparisons between the groups. The log-rank method was used to analyze the selected evaluation factors, and the significant evaluation factors in the single factor analysis were included in the multivariate factor analysis. The Cox regression model was used for the multivariate survival analysis, and a bilateral 95% CI was used as the statistical standard. The statistical analysis was performed using SPSS 20.0 (New York, IBM). Statistical significance was set at $P<0.05$.

Results

Clinicopathological features

In total, we identified 31 patients with parotid lymphoma. Among them, there were 10 males and 21 females. The age range of onset was 7-83 years old, the median age was 64 years old, and 17 cases were over 60 years old (**Table 1**). Among the 31 patients, the main clinical manifestations of the 24 cases were a painless nodule or mass in the parotid region, 9 cases had lymphadenopathy in the neck and submaxillary region, 5 cases had spontaneous pain, and 1 case had facial paralysis. The intraoperative findings indicated that there were 19 cases with tumor capsules and 12 cases without a tumor capsule. Most of the tumors were located in the superficial lobe of the parotid gland (28/31), and three cases involved the deep lobe of the parotid gland. 90.3% of the cases fell into Ann Arbor stages I or II, and 9.7% of the cases fell into Ann Arbor stages III or IV.

Morphological features

A definitive diagnosis of parotid lymphoma is often only done postoperatively because the

Table 1. Clinicopathological features of the 31 patients with parotid lymphoma

Clinical features	n (%)
Gender	
Female	21 (67.7%)
Male	10 (32.3%)
Age	
≥60 years	17 (54.8%)
<60 years	14 (45.2%)
Extent	
Unilateral	27 (87.1%)
Bilateral	4 (12.9%)
Location	
Superficial lobe	28 (90.3%)
Deep lobe	3 (9.7%)
Histologic classification	
MALT	18 (58.1%)
DLBCL	7 (22.6%)
FL	6 (19.4%)
Ann Arbor stage	
stages I and II	28 (90.3%)
stages III and IV	3 (9.7%)
Autoimmune disorders	
Sjogren's syndrome	5 (16.1%)
Rheumatoid arthritis	1 (3.2%)
Local symptoms	
Spontaneous pain	5 (16.1%)
Facial paralysis	1 (3.2%)
Accelerated tumor growth	8 (25.8%)
Tumors without capsules	12 (38.7%)
Surgery	26 (83.9%)

confirmation requires an immunohistochemical analysis, flow cytometry, cytogenetics, and a molecular diagnostic evaluation. Flow cytometry detects monoclonality in B-cells [12, 13]. An immunohistochemical analysis helps determine whether a tumor is a B-cell lymphoma (CD20+), a T-cell lymphoma (CD3+), or a carcinoma (keratin+) [14, 15]. Representative micrographs show parotid lymphoma using hematoxylin-eosin (HE) staining and immunohistochemical analysis. In MALT lymphoma, the cell nucleus is small to medium sized, slightly irregular, the cytoplasm is lightly stained, the plasma cell differentiation is more common, and scattered immunoblasts and central cells can also be seen. The tumor cells proliferate around the normal glands, and significant lymphoepithelial lesions can be seen in most cases (**Figure 1**). There was no significant difference between the morphological characteristics of DLBCL of the parotid gland and of other parts. The tumor cells were large, rich in cytoplasms, light-pink stained, and large in nuclear volume (**Figure 2**). The FL, CD10, and Bcl-2 tumor cells were positive (**Figure 3**). Among the 31 patients, there were 18 cases of extranodal marginal zone B-cell lymphoma of the mucosa-associated lymphoid tissue, 7 cases of diffuse large B cell lymphoma, and 6 cases of follicular lymphoma.

Treatment methods and outcomes

The treatment for parotid lymphoma includes local and systematic treatment, and the former includes surgical treatment and local radiotherapy. The latter includes chemotherapy and a combination of surgery with radiotherapy and chemotherapy. The chemotherapy includes rituximab-targeted therapy, conventional chemotherapy regimens, for instance, cyclophosphamide, hydroxydaunorubicin, oncovin, vincristine, prednisone (CHOP), and conventional chemotherapy regimens combined with rituximab [16]. Different treatment methods were selected according to the clinical stage, the involved site, and the patient's systemic tolerance. Among the 31 patients, 5 of them completed relevant examinations and underwent tumor resection biopsies after admission, and they were confirmed in our hospital or recommended to be transferred to other hospitals for radiotherapy, chemotherapy, or observation. 26 patients were treated with tumor resection, including 6 cases of partial parotidectomy and 20 cases of total parotidectomy. Of the 26 patients who received surgery, 6 cases were treated using surgery alone, 3 cases using radiotherapy, 14 cases using chemotherapy, and 3 cases using radiotherapy and chemotherapy. Among the 5 patients who did not undergo an operation, 4 cases only received chemotherapy, and 1 case received radiotherapy and chemotherapy. Among the 31 patients, 27 patients had a complete remission after the treatment, 3 patients had slow progress after the treatment, and 1 patient had a partial remission after the treatment.

Total survival analysis

The overall median survival from the diagnosis of lymphoma was estimated to be 62 months

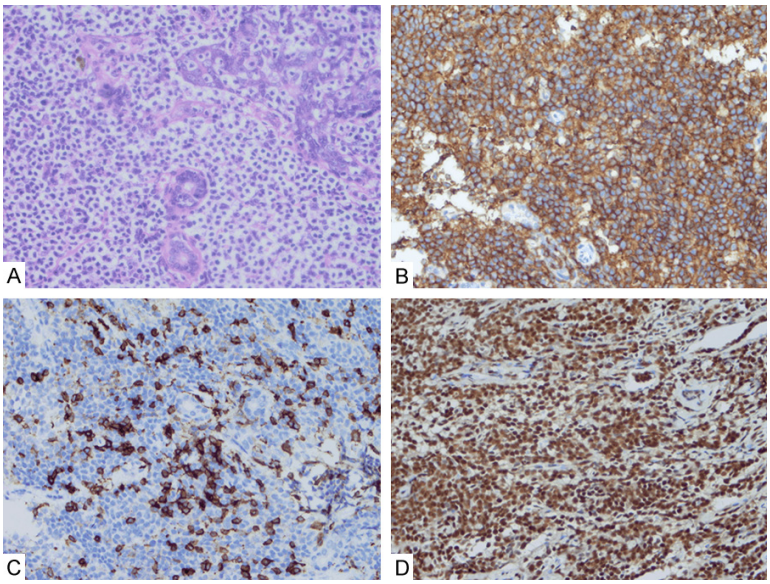

Figure 1. Histological features of lymphoma of the mucosa-associated lymphoid tissue of the salivary gland (Hematoxylin-eosin staining). (A) The normal structure of the parotid gland is destroyed by the diffuse hyperplasia of lymphoid tissue, forming lymphoepithelial lesions (magnification ×400). Immunohistochemistry for the mucosa-associated lymphoid tissue lymphoma. The tumor cells are positive for CD20 (B), CD43 (C), PAX-5 (D) (magnification ×400).

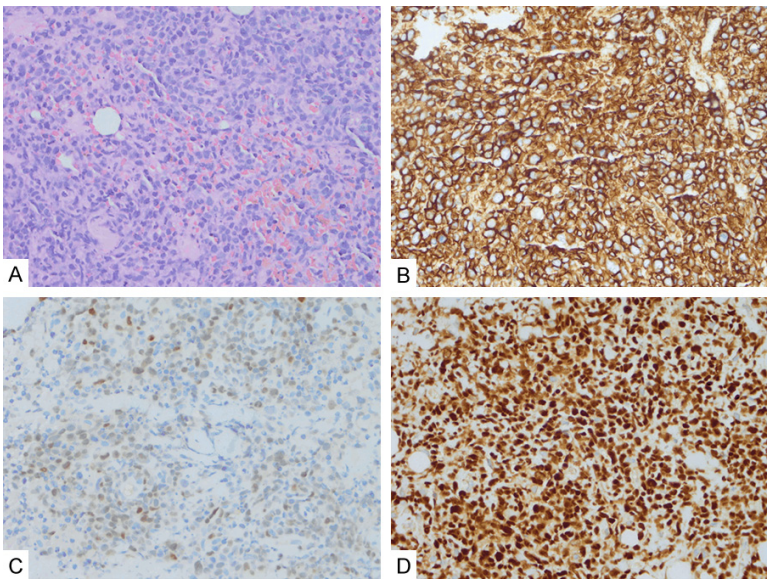

Figure 2. Histological features of diffuse large B-cell lymphoma of the salivary gland (Hematoxylin-eosin staining). (A) The tumor cells were large, rich in cytoplasms, and large in nuclear volume (magnification ×400). Immunohistochemistry for diffuse large B cell lymphoma. The tumor cells are positive for CD20 (B), MUM-1 (C), PAX-5 (D) (magnification ×400).

(the range was 29-92 months), and the mean follow-up time was (62±16) months. Among the 31 patients with parotid lymphoma, 7 died during the observation period, 5 died from ineffective treatment due to lymphoma progression, 1 died from a cerebrovascular accident, and 1 died from a traffic accident. The total 3-year survival rate was 83.9%, and the cumulative 5-year survival rate was 77.4% (**Figure 4**).

Factors affecting prognosis

A Kaplan Meier single-factor analysis showed that accelerated tumor growth ($P<0.001$) and the absence of a tumor capsule ($P<0.001$) were the factors influencing the overall survival rate. The 3-year and 5-year overall survival rates of the different groups are shown in **Table 2**. Gender, clinical-stage, pathological classification, the presence of an immune disease, whether there were local symptoms, and whether a patient underwent surgery or not had no significant effect on the overall survival of patients. Group factors with statistical significance in the single factor analysis were included in the Cox multivariate factor analysis, and the results showed that the existence of accelerated tumor growth is the factor that most affects the overall patient survival rate (**Table 3**). The overall patient survival rate of patients with accelerated tumor growth is significantly higher than it is in patients without accelerated tumor growth.

Discussion

Considering the low incidence of lymphoma of the salivary gland, the clinical manifestations of oral and maxillofacial lymphoma are diverse, the diagnostic process is relatively complex, and there are often misdiagnoses or

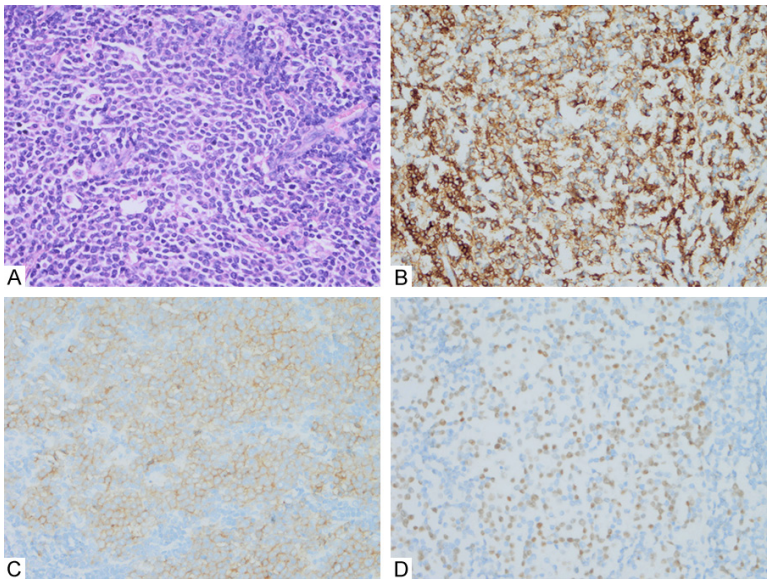

Figure 3. Representative micrographs showing follicular lymphoma (Hematoxylin-eosin stain for A, ×400). Immunohistochemistry for follicular lymphoma. The tumor cells are positive for CD20 (B), CD10 (C), Bcl-6 (D) (magnification ×400).

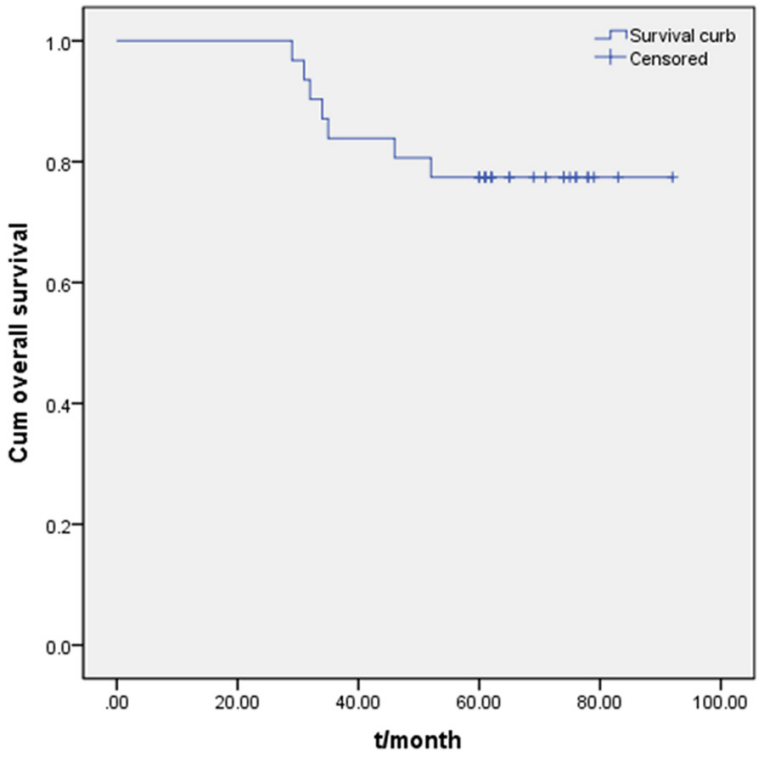

Figure 4. The overall survival curve for patients with parotid lymphoma.

delayed diagnoses [17]. The definitive diagnosis depends on the pathological results of the postoperative specimens. Lymphoma of the parotid gland is difficult to differentiate from other tumors of the parotid gland, so it is impossible to make a definite diagnosis preoperatively, so an operation or a biopsy is needed to make a definite diagnosis. In a majority of lymphoma of the parotid gland cases, the parotidectomy is more likely to be more diagnostic than therapeutic. At present, there is no unified standard for the definitions of primary and secondary parotid lymphoma. After reviewing and consulting the relevant literature, we think the diagnosis of primary parotid lymphoma should meet the following standards [18], (1) The first site or symptom of the tumor was found in the parotid gland, and the tumor was shown to be involved in the parenchyma of the parotid gland using pathology. (2) There is no superficial lymphadenopathy (except in the regional lymph nodes). (3) There is no hepatosplenomegaly or mediastinal lymphadenopathy. (4) The peripheral hemogram and the myelogram are normal. (5) It was 6 months after the first malignant lymphoma appeared that malignant lymphoma appeared in other parts. The clinical symptoms of primary lymphoma of the parotid gland are mostly painless masses in front of the ear, but the masses may be accompanied by pain in the later stages. It is generally believed that spontaneous pain, facial paralysis, and accelerated tumor growth are typical manifestations of parotid gland cancer [4].

Immunohistochemistry proved to be useful in obtaining a correct diagnosis, and it is also suitable for diagnosing adenolymphoma, pleomor-

Table 2. Analysis of the risk factors associated with survival for patients with primary parotid lymphoma according to a univariate analysis

Risk factor	3-year OS/%	5-year OS/%	X^2	P
Gender			2.510	0.113
Female	80.9	76.2		
Male	90.0	80.0		
Age			0.018	0.895
≥60 years	76.5	64.7		
<60 years	92.9	92.9		
Location			0.004	0.950
Superficial lobe	85.7	82.1		
Deep lobe	66.7	33.3		
Accelerated tumor growth			12.351	0.000
Yes	50.0	50.0		
No	95.7	86.9		
Ann Arbor stage			0.249	0.618
stages I and stage II	85.7	78.6		
stages III and stage IV	66.7	66.7		
Histologic classification			1.273	0.259
MALT	88.9	77.8		
Other subtypes	76.9	76.9		
Local symptoms			2.548	0.110
Yes	75.0	62.5		
No	86.9	82.6		
Surgery			0.407	0.523
Yes	84.6	80.8		
No	80.0	60.0		
Tumor capsule			25.884	0.000
Yes	94.7	89.5		
No	66.7	58.3		

OS, overall survival.

Table 3. Analysis of the risk factors associated with survival for patients with primary parotid lymphoma according to a multivariate analysis

Risk factor	HR (95% CI)	P
Accelerated tumor growth (Yes/No)	4.790 (0.041-0.839)	0.029

phic adenoma, carcinoma of the parotid gland, and metastatic tumors. The majority of primary lymphoma of the parotid gland is B cell NHL, accounting for 84-97%, and the minority is T cell NHL or HL [19]. In this study, 31 patients were B-cell NHL. Considering the small number of samples, there is a certain degree of randomness. Extranodal marginal zone B-cell lymphoma of the mucosa-associated lymphoid tissue (MALT) and follicular lymphoma (FL) are the most common histological classifications of parotid lymphoma [10]. Inconsistent with previous published studies, our study finds that MALT lymphoma is the most common, accounting for 58.06% (18/31), DLBCL is the second most common, accounting for 22.58% (7/31), followed by FL, accounting for 19.35% (6/31). The difference between the results may be due to the small sample size. Owing to the fact that primary lymphoma of the parotid gland is mainly MALT, which is low-grade lymphoma, the growth of primary lymphoma of the parotid gland is mostly slow. DLBCL is the most common NHL, and it can be transformed from low-grade lymphoma such as MALT lymphoma or FL [6, 20]. Therefore, we should pay more attention to the patients who have a history of accelerated tumor growth. Studies state that when the Ki-67 proliferation index is less than 10%, the cases tend to have shorter survival times when compared with high proliferation activity, but the difference was not statistically significant [11]. However, the study reports that a high Ki-67 proliferation index is a marker of poor prognosis for DLBCL [21]. To some extent, the Ki-67 expression level can reflect the prognosis of lymphoma [22].

There is no unified plan for the treatment of parotid lymphoma. Surgery is a common treatment, but some scholars do not recommend it as the first-line treatment for parotid lymphoma [20]. Relevant research shows that the main diagnostic tools for parotid lymphoma are tumor resection and superficial lobectomy [23]. The treatment for MALT lymphoma of the parotid gland is different from the treatment for other non-Hodgkin's lymphomas. Because most parotid MALT lymphoma cases involve localized masses and have a low malignancy and less systemic metastasis, its treatment is mainly surgical resection with further radiotherapy and chemotherapy after the operation, and

the prognosis is good. The analysis of the prognostic factors in this study showed that the surgical factors had no significant effect on the prognosis of parotid lymphoma. One study found that adjuvant local radiotherapy can control tumor development [24]. In recent years, new chemotherapy drugs, especially biologically targeted drugs, have achieved remarkable results in the field of lymphoma treatment [25]. A monoclonal antibody targeting CD20, rituximab significantly improves the survival rate of B-cell lymphoma patients and plays an important role in the treatment of parotid lymphoma [26]. In this study, we described many methods of treatment, most of which are surgery combined with chemotherapy. According to the results of this study and the medical literature, conservative surgery combined with postoperative chemotherapy can be used for parotid lymphoma. When clinical symptoms and signs, as well as the results of the auxiliary examination, suggest that parotid lymphoma tumors and their surrounding parts should be removed, a frozen section examination should be performed during the operation to preliminarily determine the diagnosis of lymphoma. Postoperative chemotherapy was carried out according to the pathological type and molecular phenotype of the tumor. There is not only local treatment for the resection of the main lesions, but there is also systemic treatment for the whole body. Sufficient tissue samples can be obtained during operations, and complications such as radiation dry mouth caused by local radiotherapy can be avoided.

In this study, the 3-year survival rate of the 31 patients with primary parotid lymphoma was 83.9%, and the 5-year survival rate was 77.4%. Compared with other types of parotid cancer, the prognoses were relatively good. The international prognostic index (IPI) is widely used to evaluate the prognosis of lymphoma. It mainly includes patients' ages and clinical stages, their level of LDH in the concomitant serum, their Eastern Cooperative Oncology Group (ECOG) scores, the presence of B symptoms, and the invasion of their extranodal tissues [27]. The results of the single-factor analysis showed that accelerated tumor growth and a lack of tumor capsules were the prognosis risk factors, and the clinical-stage had no significant effect on prognosis. The prognosis of primary parotid lymphoma may be different from that of other parts. For example, all primary parotid lymphoma is an extranodal invasion, which may overestimate the IPI score. Therefore, the role of IPI in the prognosis evaluation of parotid lymphoma remains to be determined.

Conclusion

In summary, primary parotid lymphoma is a rare malignant tumor of the salivary gland. The diagnosis of this disease includes its clinical manifestations, immunohistochemical staining, and a cytogenetic analysis. The therapeutic method is primary surgical excision and chemotherapy or radiotherapy, according to the clinic. This study aimed to improve the awareness of its diagnosis and to review the current views on this rare neoplasm. In additional, patients should be followed up for a longer time to guide their care and prognosis.

Disclosure of conflict of interest

None.

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