THE IMMUNOHISTOCHEMISTRY AND QUANTITATIVE MORPHOMETRIC STUDY OF MUCOSA ASSOCIATED LYMPHOID TISSUE (MALT) LYMPHOMA IN STOMACH

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ABSTRACT

Objective: To invetigate the Immunohistochemistry characters and quantitative nmorphometric significance for the mucosa associated lymphoid tissue (MALT) lymphoma of stomach in 14 patients. Methods: The routine paraffin slides were cut, stained with H.E., and immunochemically by ABC method. The morphologic appearance of nuclei of lymphoma cells were measured with HPIAS-1000 color pathology picture analysis system. Results of the 14 cases, 9 was centrocyte like (CCL) cell lymphoma, 2 CCL with large cell lymphoma, 1 small no cleaved cell lymphoma, 1 large no cleaved cell lymphoma, 1 T immunoblastic malignant lymphoma. The morphologic measurement results showed that there were great significant differences ($P<0.001$) for the 15 items of morphology parameters between the nuclei of MALT lymphoma cells and those of normal control lymphocytes in stomach. There were great significance differences ($P<0.001$) or significance ($P<0.05$) for the most of the 15 items of morphologic parameters of nuclei among the 5 types of MALT lymphoma. Especially, that the values of area, circumference, equivalent diameter, area volume, circumference volume, long diameter, short diameter, practical area were increasing as the malignant degree of classification was rising, which reflect the increasing malignancy of the tumor. Conclusion: It was suggested that with the quantitative morphology measurement method, man could make accurate diagnosis for MALT lymphoma. It offered us a new method to make the diagnosis, so that it had significance. It might be also practicable with morphology measurement method to make the sub classification of MALT lymphoma.

Key words: Stomach, MALT lymphoma, Pathologic morphology, Morphometry

The mucosa associated lymphoid tiuuse (MALT) lymphoma is a sort of lower malignant lymphom originated from the marginal zone of lymphatic follicles with special pathological morphology and clinical characters.[1-3] Although in past tine, much research work for MALT has been done, however, as the morphology and the types of MALT lymphoma are very complicated, it is now still sometime very difficult for us to make correct diagnosis and differential diagnosis. In this article we report 14 cases of primary MALT quantitative morphology parameters, and probe their significance for the diagnosis and differential diagnosis. We have not seen any similar report in China before.

MATERIALS AND METHODS

Fourteen cases with primary MALT lymphoma of stomachs were collected from 1994 to 1998. The samples were fixed with 3% methanol. The routine paraffin slides were cut and stained with H.E and immunochemistry ABC methods. The antibodies used were offered by DAKO company. The slides were observed under optical microscope. In addition, the quantitative parameters of nuclei of MALT lymphoma in slides with typical morphology were measured with HPIAS-1000 color picture morphology analysis system of Qing Ping Image Company. The objective (40X) was used for measurement. For each case at least 300 lymphoma cells were measured. The lymphocytes in normal stomach mucosa of 24 healthy

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persons were selected as control.

RESULTS

Clinical Data

Of the 14 cases, 8 were male, 6 were female. The rate of male to female was 1.3: 1. The ages ranged between 27 to 78. The mean age was 57. The clinical symptoms were upper abdomen pain and upset (12 cases, 85.7%), vomit or hematemesis (7 cases, 50.0%), belch and vomiting of acid (5 cases, 35.7%), inappetence and obvious liposarcous (4 cases, 28.5%), black stool and hemafecia (3 cases, 21.5%), and ascites (1 case, 7.1%) etc.. There were no signs of hepatomegaly, splenauxe, and lymphadenovarix for all cases.

Gross Examination

The neoplasms localized in antrum (6 cases), small curvature (4 cases), great curvature (3 cases) and in the rear wall of stomach (1 case). The largest one of the tumor was 9 cm×6 cm×2.5 cm, the smallest one was 2.5 cm×2.5 cm×1cm. The common size of the tumors was about 4 cm×2.5 cm×2 cm (7 cases). The surface of the tumor was ruffle and coarse, and in 12 cases, the superficial ulcers could be seen. The bottom of the ulcers was ruffle with necrotic tissue. The cutting surface of the tumors was white and pink. The texture was fish flesh like.

Microscope Examination

There was diffusely dense infiltration of lymphoma cells in the mucosa for all cases, in submucosa for 8 cases and in muscular layer for 6 cases. There were some remained atrophic glands sparsely distributed in the tumor. In 10 cases, the lymphoma cells were infiltrating among epithelia (lymph-epithelial disease). In 5 cases, the plasma transformation of lymphoma cells, in 10 cases, the necrosis and in 2 cases, the remained lymphatic follicles could be seen. In the tumors, there were often some mild (11 cases) or remarkable (3 cases) proliferative capillaries. The mitoses were usually rare. Of the 14 cases, 12 cases had mitoses of 0~1/10 HPF, 2 cases had mitoses of 2~6/10 HPF in the tumors. The histology types were centrocyte like (CCL) cell lymphoma (9 cases), CCL with large cell lymphoma (CCLWLCL, 2 cases), small no cleaved cell lymphoma (SNCCL, 1 case), large no cleaved cell lymphoma (lNCCL, 1 case) and T immunoblastic malignant lymphoma (T IBML, 1 case).

Immunochemistry

For most (11 cases) of the 14 cases, the lymphoma cells expressed LCA(+). There were often some Mac_{387} positive cells sparsely distributed among the lymphoma cells (6 cases). Of the 14 cases, 13 cases (92.8%) were B cell lymphoma. Among them, 12 cases expressed L_{26} (+), 2 cases IgG (+), 1 case IgM (+), 1 case IgA (+). Seven cases expressed κ (+), and 4 cases λ (+). One case of T IBML expressed UCHL-1(+).

Morphometry Measurement

The morphometry measurement results of 14 cases were listed in Table 1. The statistic examination results for the difference of parameters of each type of lymphomas were listed in Table 2.

DISCUSSION

Although the primary malignant lymphomas of gastrointestinal tract (PMLGI) were rare, they were the most common encountered lymphomas in extra lymph node sites. The dominant type of PMLGI was MALT lymphoma. It was suggested by WHO that it originated from the marginal zone of lymphatic follicles. Generally, the tumors were located in antrum, prevalently in old patients with superficial ulcers in the surface. Microscopically, the characters of it were CCL cells with plasma transformation, lymph-epithelial disease and remained lymphatic follicles among the lymphoma cells.[4] The neoplasms usually developed slowly, and the patient's prognoses were relatively well. In our group, the main type of the tumors was CCL lymphoma, the clinical symptoms, the characters of gross examination and morphology under microscopy were consistent with those depicted in literature. It was reported that the low malignant MALT lymphoma might show the transformation of metrocyte, namely, sheets of centroblastic like cells or immunoblastic like cells occurring among the lymphoma cells. Some people called it high malignant MALT lymphoma. There were 2 cases of CCLWLCL in our group, consistent with the high malignant lymphoma. It was still in controversy whether the pure large B cell lymphoma was primary MALT lymphoma.[5] We had only one such case in our group. It was rare.

Over a long period of time, a hard nut for pathologists to crack was how to make correct diagnosis and differential diagnosis for PMLGI. Especially, the CCL lymphoma cells were so similar to the lymphocytes of marginal zone of lymphatic

follicles that man could hardly discriminate them, so came a name of Pseudolymphoma. The key for differential diagnosis of classic pathology was recognition of remained lymphatic follicles and examination with monoclonal antibodies.[4] However, the Hyperplastic or atrophic lymphatic follicles could frequently be seen in chronic inflammation, and the immunochemistry examinations often had pseudo positive or pseudo negative results because of unsuitable fixation of tissue and insufficient purification and specialty of the antibodies. The morphologic measurement results of our 14 cases showed that there were great significant differences ($P<0.001$) for the 15 items of morphology parameters between the nuclei of MALT lymphoma cells and those of normal control lymphocytes in stomachs. It suggested that with the quantitative morphology measurement method, man could make accurate diagnosis for MALT lymphoma. It offered us a new method to make the diagnosis, so that it had significance.

The pathologic classifications of MALT lymphoma in stomach are important to the treatment and the assessment of the patients' prognosis.[6] Through many years consecutively hard working of pathologists', the classifications have basically been defined. However, the most obvious defeats of the classifications under microscope with naked eyes are the subjectivity and the arbitrariness, so the foundation of a more objective and accurate classification is a crucial problem to be resolved. Although all the existed classifications of lymphomas in world are complicated, the common feature of them is that they are nominated according to the different stages of transformation of lymphocytes, and each stage of them has the specific morphologic features, so the classification through the morphometry measurement has the objective morphologic basis and is theoretically practicable. The morphologic measurement results of our 14 cases showed that there was great significance ($P<0.001$) or significance ($P<0.05$) for the most of the 15 items of morphologic parameters of nuclei among the 5 types of MALT lymphoma, especially that the values of area, circumference, equivalent diameter, area volume, circumference volume, long diameter, short diameter, practical area were increasing from CCL cell lymphoma, SNCCL, CCLWLCL to LNCCL as the malignant degree of classification was rising, which reflect the increasing malignancy of the tumor. The morphologic parameters of 1 case of T IBML were similar to those of LNCCL, which reflected both of them were high malignant. Our measurement results suggested that it might be practicable with morphology measurement method to make sub classification of MALT lymphoma. We believe that along with the gradually popularization and application of quantitative morphologic measurement methods its importance will more and more be attached.

Table 1. The quantitative morphologic parameters of nucleus of MALT lymphoma of stomach in 14 cases ($\bar{x}\pm s$)

	CCL	CCLWLCL	SNCCL	LNCCL	T IBML	Control
Cases	9	2	1	1	1	24
Cells	3072	1029	491	625	681	1738
Area	130613± 0.247	17.685± 0.393	18.570± 0.449	26.159± 0.655	21.574± 0.782	12.123± 0.172
Cir	130754± 0.137	15.771± 0.225	16.060± 0.235	19.460± 0.343	17.342±0.397	12.796± 0.094
ED	40155± 0.123	4.722± 0.196	4.832± 0.221	5.690± 0.331	5.153± 0.364	3.925± 0.086
AV	38.007± 0.372	56.786± 0.595	61.364± 0.686	104.813± 0.972	79.577± 1.254	31.846± 0.257
CV	45.013± 0.414	69.555± 0.689	73.725± 0.731	137.303± 1.011	100.719± 1.368	35.960± 0.274
LD	40643± 0.145	5.305± 0.219	5.390± 0.247	6.502± 0.339	5.845± 0.370	4.279± 0.110
SD	30787± 0.155	4.277± 0.221	4.407± 0.225	5.084± 0.355	4.627± 0.393	3.666± 0.108
LD/SD	10231± 0.174	1.247± 0.209	1.227± 0.176	1.289± 0.220	1.275± 0.243	1.170± 0.135
MF	1.110± 0.094	1.129± 0.113	1.118± 0.078	1.186± 0.141	1.143± 0.133	1.077± 0.053
RD	0.903± 0.092	0.889± 0.111	0.896± 0.076	0.847± 0.133	0.879± 0.127	0.929± 0.053
ED	0.009± 1.470	0.010± 1.671	0.008± 1.507	0.012± 1.495	0.013±1.779	0.005± 1.511
RD	0.861± 0.098	0.849± 0.105	0.840± 0.086	0.813± 0.106	0.833± 0.106	0.877± 0.086
GD	6.681± 0.092	6.579± 0.111	6.629± 0.076	6.272± 0.133	6.503± 0.127	6.879± 0.053
AI	3.734± 0.047	3.765± 0.056	3.748± 0.039	3.858± 0.071	3.788± 0.066	3.678± 0.027
PA	13.667± 0.247	17.732± 0.395	18.648± 0.449	26.225± 0.653	21.636± 0.774	12.167± 0.171

Notes: Cir: circumference. ED: equivalent diameter. AV: area volume. CV: circumference volume. LD: long diameter. SD: short diameter. LD/SD: long diameter to short diameter. MF: morphologic factor. RD: round degree. ED: ellipse degree. RD: rectangle degree. GD: globe degree. AI: atypical indicator. PA: practical area.

Table 2-1. The remarkable significant differences of nuclear morphologic parameters of respective types of MALT lymphoma in stomachs (U value)

	CCL /control	CCLWLCL/ control	SNCCL/ control	LNCCL/ control	T IBML/ control	CCL/ CCLWLCL	CCL/ SNCCL	CCL/ LNCCL
Area	37.157	48.823	33.253	40.103	28.589	35.347	25.460	35.795
Cir	42.239	51.038	37.104	48.642	33.515	3.417	26.071	41.559
ED	36.566	52.126	36.426	45.523	33.270	36.818	27.083	39.526
AV	37.595	45.531	30.334	72.967	24.438	33.898	23.917	32.089
CV	43.013	43.528	30.304	35.718	24.004	31.416	22.933	32.498
LD	43.230	50.997	35.624	48.976	36.592	33.914	4.197	40.902
SD	16.621	38.549	31.613	38.245	26.665	30.625	4.047	8.674
LD/SD	22.206	16.853	10.710	19.605	16.532	3.487	0.750**	9.530
MF	27.966	23.941	19.105	31.071	21.509	8.559	3.571	16.594
RD	26.918	23.955	19.538	34.224	22.076	8.092	3.998	22.951
ED	25.823	16.892	9.130	17.032	16.466	3.279*	2.962	7.177
RD	13.223	16.355	19.331	32.112	22.449	7.362	11.407	25.000
GD	27.627	24.106	13.646	34.333	22.475	7.907	3.985	22.722
AI	28.000	24.147	19.493	32.143	21.786	8.251	2.297*	21.754
PA	37.406	48.675	33.407	40.194	28.834	35.171	25.570	35.870

Table 2-2

	CCL/ T IBML	CCLWLCL/ SNCCL	CCLWLCL/ LNCCL	CCLWLCL/ T IBML	SNCCL/ LNCCL	SNCCL/ T IBML	LNCCL/ T IBML
Area	24.048	3.988	23.128	11.182	19.044	7.873	9.550
Cir	26.401	2.792*	26.927	10.748	21.053	7.996	11.050
ED	26.988	3.846	23.461	100.906	18.775	7.274	10.094
AV	21.265	4.133	22.375	11.263	18.954	8.364	8.855
CV	20.628	2.865	23.080	11.127	20.548	9.099	9.353
LD	28.045	2.374*	24.584	11.668	20.404	8.696	10.620
SD	7.535	4.745	20.327	9.028	15.628	5.184	8.917
LD/SD	5.070	3.092	5.919	3.816	8.158	6.131	1.675**
MF	10.516	3.806	14.286	3.859	17.043	6.919	9.438
RD	10.390	3.156*	15.027	3.727	17.438	6.308	10.047
ED	9.132	4.751	4.099	5.882	7.859	9.524	1.724**
RD	14.815	4.091	15.859	7.159	11.129	2.917*	8.130
GD	10.421	3.022*	14.838	3.822	17.180	6.309	9.830
AI	10.475	3.545	14.419	3.852	17.083	6.711	9.517
PA	24.232	4.121	23.192	11.289	19.023	7.869	13.283

Notes: Without * means $P<0.001$; * means $P<0.05$; ** means $P>0.05$.

REFERENCES

[1] Isaacson PG, Wright DH. Extranodal malignant lymphoma arising from mucosa-associated lymphoid tissue. Cancer 1984; 53: 2515.

[2] Zhang YJ, Gao XL, Li YL. The immunochemistry probe of five cases of MALT lymphoma. Chin J Pathol 1995; 6: 175.

[3] Zhang YJ, Li YL, Zhu GB, et al. Pathological study of gastrointestinal mucosa-associated lymphoid tissue-derived lymphoma. J Clin Exp Pathol 1994; 10: 119.

[4] Cai RX, Zhang HZ, Zhou JH, et al. New classificatory pathomorphological study on 62 cases with primary malignant lymphoma of gastrointestinal tract. J Clin Exp Pathol 1999; 15: 5.

[5] Chan JKC, Ng CS, Isaacson PG. Relationship between high-grade lymphoma and low-grade, B-cell mucosa-associate lymphoma tissue lymphoma of the stomach. Am J Pathol 1990; 136: 1153.

[6] Pan WS, Xu YD. Morphometrical study and classification of malignant lymphoma. J Clin Exp Pathol 1991; 7: 161.