

# Histopathological Difference of Nasopharyngeal Carcinoma in Southern China and Japan

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Histopathological analysis was carried out to examine the characteristics of nasopharyngeal carcinoma (NPC) using 38 biopsy cases obtained from southern China and 34 cases obtained from Japan. These cases were divided into three groups according to their predominant pattern associated with the cell and the tissue differentiation using the World Health Organization (WHO) histological classification of tumors as follows: (i) squamous cell carcinoma; 6 Chinese cases and 5 Japanese cases, (ii) differentiated non-keratinizing carcinoma; 25 Chinese cases and 7 Japanese cases, (iii) undifferentiated carcinoma; 7 Chinese cases and 22 Japanese cases. The age of Chinese patients ranged from 21 to 80 years with a mean of 46 years and Japanese patients ranged from 17 to 89 years with a mean of 53.8 years. The male to female ratio was 2.7:1.0 in China and 1.8:1.0 in Japan. The most common histological type of NPC in China was differentiated non-keratinizing carcinoma, however, it was undifferentiated carcinoma in Japan. The mechanism of cancer arising from the nasopharynx may be quite different between Chinese and Japanese cases. The Chinese NPC cases were considered as continuity shift from normal epithelium to differentiated non-keratinizing carcinoma, to undifferentiated carcinoma sequence. On the other hand, the Japanese cases were considered discontinuity shift. Therefore, in Chinese cases, NPC formation mechanism may be that EBV infection and chemical carcinogens can act synergistically in the enhancement of both viral replication and malignant transformation. It is suggested that some Japanese cases are associated with Epstein-Barr virus infection, although, mechanism of formation of NPC in Japanese cases is not yet clear.

## Introduction

Nasopharyngeal carcinoma (NPC) was first reported as a neoplasm occurring in the nasopharynx, characteristic by anaplastic cells surrounded by prominent infiltration of lymphoid cells, this tumor has been designated as "lymphoepithelioma" or NPC. The nucleus features of the carcinoma cells are distinctive with large nucleus and prominent nucleolus. The cytoplasmic features of the carcinoma cells are variable and the cellular borders are often indistinct due to fusion of cytoplasm. NPC has been

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confirmed as a tumor arising from squamous epithelium by electron microscopical and immunohistochemical examination. According to the World Health Organization (WHO) histological classification, NPC was divided into three types; squamous cell carcinoma, differentiated non-keratinizing carcinoma, and undifferentiated carcinoma.<sup>15)</sup> The Epstein-Barr virus (EBV) is associated with a variety of benign and malignant lymphoproliferative diseases, most notably infectious mononucleosis, Burkitt's lymphoma, and NPC. The presence of EBV has been reported in undifferentiated NPC, and partially differentiated non-keratinizing NPC, but not in squamous cell NPC. Lymphoepithelioma of the nasopharynx has been shown by electronmicroscopical and immunohistochemical studies to be an anaplastic variant of squamous cell carcinoma. There is many evidences supporting the strong association between EBV and nasopharyngeal lymphoepithelioma and NPC. Lymphoepitheliomas occur outside of the nasopharynx, although they are rare. Histologically similar undifferentiated carcinoma associated with marked lymphoid cell infiltration is called lymphoepithelioma-like carcinoma which have been found in the tonsil,<sup>11)</sup> salivary gland,<sup>8, 12)</sup> thymus,<sup>19)</sup> uterine cervix,<sup>9)</sup> skin,<sup>18)</sup> lung,<sup>2)</sup> and stomach.<sup>17)</sup>

In this report, we described histopathological characteristics of NPC in China and Japan using 38 caese of Chinses patients and 34 cases of Japanese patients. This investigation was undertaken as an extention of our previous work.<sup>13, 14, 21)</sup>

## Materials and Methods

The NPC specimens from 38 biopsy cases at the Jinan University Hospital in southern China and 34 biopsy cases at the Nagasaki University Hospital in Japan were used in this study. Shanmugaratnam and Sobin criteria were applied in assigning the diagnosis of NPC to the tissue. These NPC cases were divided into three groups; (i) squamous cell carcinoma type, (ii) differentiated non-keratinizing carcinoma type, and (iii) undifferentiated carcinoma type.

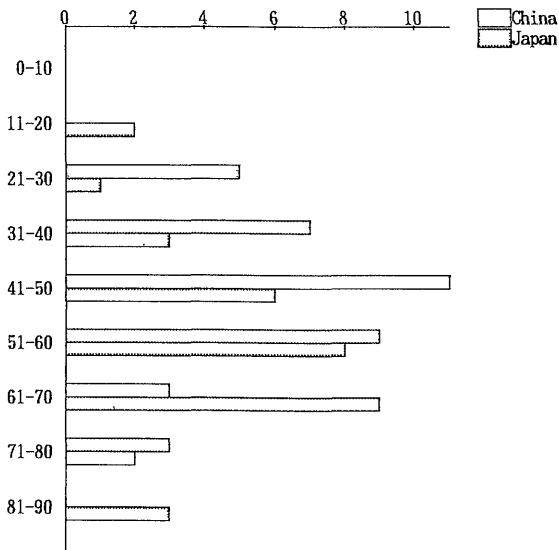
The specimens were fixed in 10% formalin, and em-

bedded in paraffin for histopathological study. Sections were cut at 4 micron and stained with hematoxylin-and-eosin for histological examination.

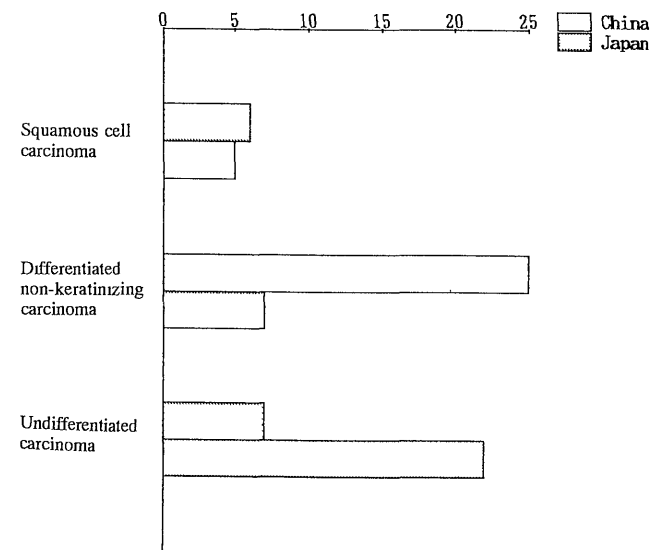
**Results**

The Chinese patients ranged in age from 21 to 80 years with a mean of 46 years. The male to female ratio was 2.7:1.0 in China. The Japanese patients ranged in age from 17 to 89 years with a mean of 53.8 years. The male to female ratio was 1.8:1.0 in Japan. Fig. 1 shows age distribution of NPC in China and Japan. The highest incidence of NPC in China was found in the 41-50-year-old age group, and NPC in Japan was detected with highest incidence in the 61-70-year-old age group. In China, no case

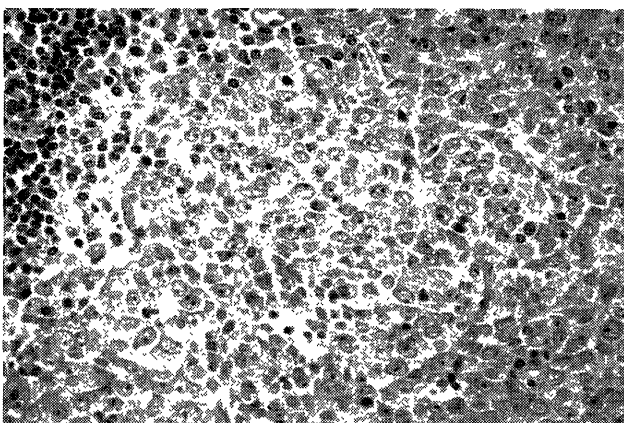
was found in the 11-20-year-old and 81-90-year-old age groups. The sex distribution revealed that males had higher rates than females in China as well as in Japan. As shown in Fig. 2, the incidence of squamous cell carcinoma in the Japanese cases was similar to the incidence of Chinese cases. The most common histological type of NPC in China was differentiated non-keratinizing carcinoma. On the other hand, NPC in Japan was undifferentiated carcinoma was higher than in China. These findings showed different incidence of histological classification of NPC in different geographic groups. Histologically, squamous cell carcinoma was quite different between China and Japan, because most of the Chinese cases were moderately differentiated squamous cell carcinoma (Fig. 3), but most of the Japanese cases were well differentiated squamous cell carcinoma (Fig. 4). Differentiated non-keratinizing



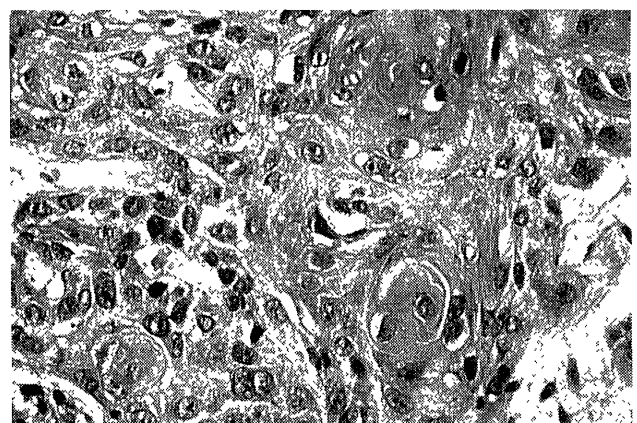
**Fig. 1.** Age distribution of nasopharyngeal carcinoma in China and Japan.



**Fig. 2.** Distribution histological classification of nasopharyngeal carcinoma in China and Japan.

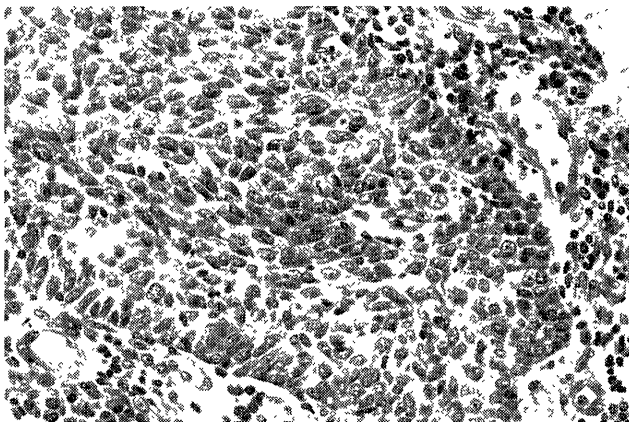


**Fig. 3.** China case: Squamous cell carcinoma type of nasopharyngeal carcinoma with abundant lymphoid cell infiltrate. Hematoxylin and eosin stain, X 200.

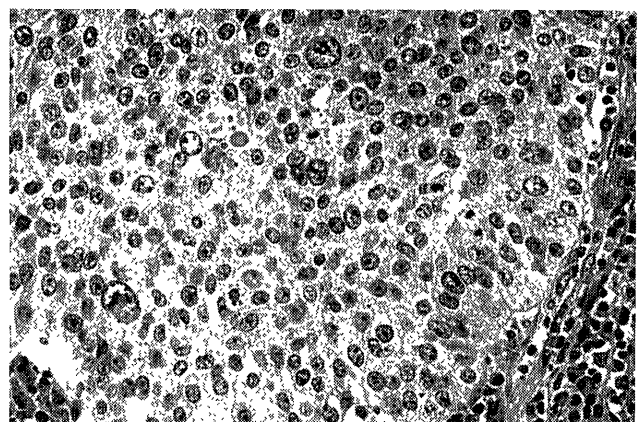


**Fig. 4.** Japan case: Squamous cell carcinoma type of nasopharyngeal carcinoma with numerous lymphoid cell infiltrate. Hematoxylin and eosin stain, X 200.

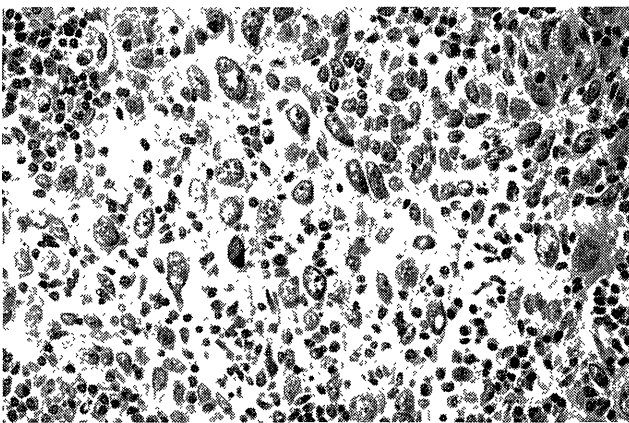
carcinoma type was similar in China and Japan (Figs. 5 and 6). Most of the undifferentiated carcinoma in China showed pleomorphism in the cytoplasm type (Fig. 7). On the other hand, most of the undifferentiated carcinoma in Japan showed the lymphoepithelioma type (Fig. 8). Summary of histogenesis of NPC in China and Japan is shown in Fig. 9. Chinese cases had continuity shift, on the other hand, Japanese cases has shown discontinuity shift. Because, in Chinese cases, coexistence of squamous cell carcinoma and differentiated non-keratinizing carcinoma were seen in the same tumor tissue, and also coexistence of differentiated non-keratinizing carcinoma and undifferentiated carcinoma were demonstrated in the same neoplastic tissue. However, such phenomena were not detected in the same cancerous tissues in Japanese cases.



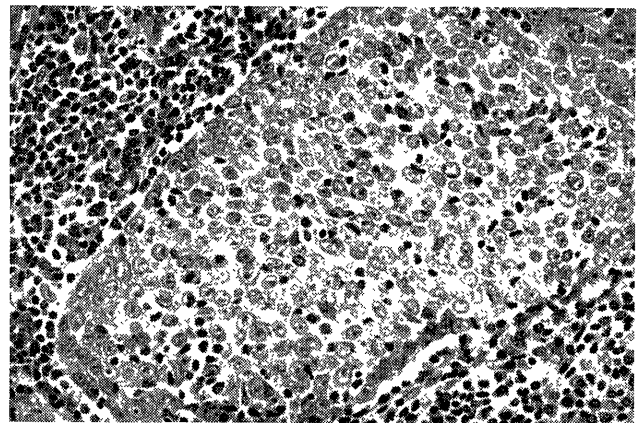
**Fig. 5.** China case: Differentiated non-keratinizing carcinoma type of nasopharyngeal carcinoma with heavy lymphoid cell infiltrate. Hematoxylin and eosin stain, X 200.



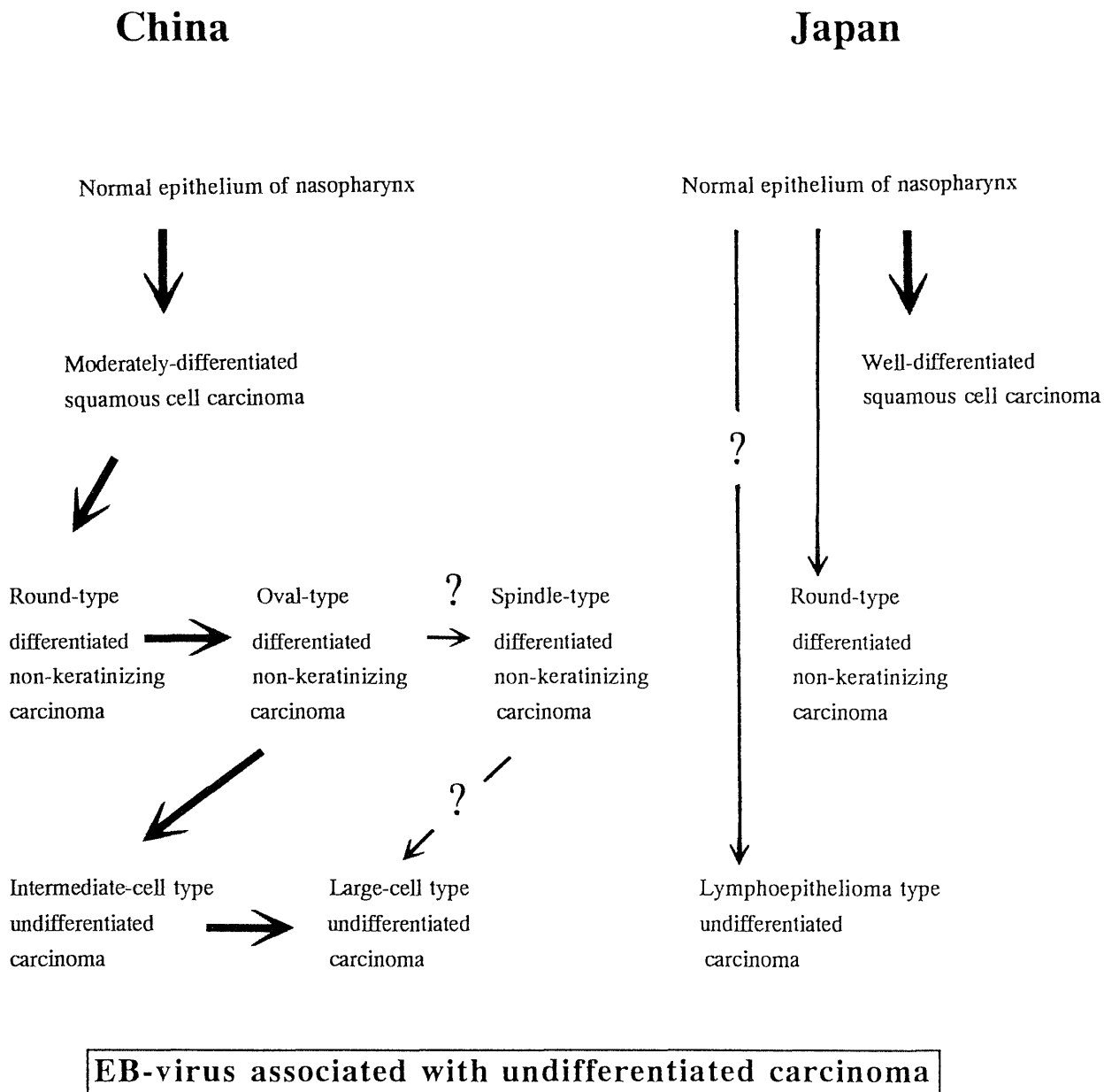
**Fig. 6.** Japan case: Differentiated non-keratinizing carcinoma type of nasopharyngeal carcinoma with abundant lymphoid cell infiltrate. Hematoxylin and eosin stain, X 200.



**Fig. 7.** China case: Undifferentiated carcinoma (non-lymphoepithelioma) type of nasopharyngeal carcinoma with numerous lymphoid cell infiltrate. Hematoxylin and eosin stain, X 200.



**Fig. 8.** Japan case: Undifferentiated carcinoma (lymphoepithelioma) type of nasopharyngeal carcinoma with heavy lymphoid cell infiltrate. Hematoxylin and eosin stain, X 200.



**Fig. 9.** Histogenesis of nasopharyngeal carcinoma in China and Japan. The authors conclude that Chinese cases are continuity shift, on the other hand, Japanese cases are discontinuity shift.

**Discussion**

Thirty-eight cases of NPC collected from southern China and 34 cases of NPC collected from Japan were examined. Malignant neoplasms primarily arising from the nasopharynx are most commonly carcinoma or malignant lymphoma. Undifferentiated NPC is accompanied by prominent lymphocytic infiltration. Therefore, it is often difficult to differentiate from malignant lymphomas, especially of the large-cell type. The typical histological appearance is undifferentiated carcinoma with infiltration of

lymphoid cells that may be so prominent as to suggest a malignant lymphoma. Immunohistochemical techniques have been proven useful in the diagnosis of nasopharyngeal lymphoepithelioma and NPC. NPC is a definite malignant epithelial neoplasm and can be distinguished from malignant lymphoma by immunohistochemical staining for keratin, epithelial membrane antigen (EMA), and carcinoembryonic antigen (CEA).<sup>21)</sup> Specifically, keratin is the most reliable epithelial marker for identifying NPC and excluding malignant lymphoma.<sup>16)</sup> In general, nasopharyngeal lymphomas have been studied under the category

of Waldeyer's ring lymphoma or nasal lymphoma. Many researchers reported that most of Waldeyer's ring lymphoma cases were B-cell phenotype. On the other hand, most of nasal lymphoma cases were T-cell phenotype.<sup>3,5)</sup> The incidence of malignant lymphoma per 100,000 population was 11-32 in Japan, and 0.1-0.9 in China. Therefore, malignant lymphoma in nasopharynx is more frequent in Japan than in China. In the nasopharynx, a component of Waldeyer's ring has abundant lymphoid tissues, and nasopharyngeal lymphoma is more frequent in Japan than in China.

Epstein-Barr virus (EBV), a human herpesvirus, is strongly linked with two forms of B-cell lymphoma and with a much more prevalent epithelial malignancy, undifferentiated NPC and partially differentiated non-keratinizing NPC.<sup>4,10,20)</sup> EBV has been closely associated with the undifferentiated NPC, which is particularly common in the high risk area in southern China. However, EBV normally infects pharyngeal epithelial cells and is often found in the salivary gland tissue. Thus, the presence of EBV positive tumors in the nasopharynx and salivary gland is not surprising. Current methods for detection of EBV are as follows: southern blot analysis, DNA amplification in situ hybridization with either isotopic probes or nonisotopic probes, and immunohistochemistry with anti-latent membrane protein (LMP), terminal protein (TP or LPM2), and six nuclear antigens (EBNAs 1, 2, 3a, 3b, 3c, EBNA-leader protein). As to NPC formation in southern China three interacting factors considered: (i) abnormal HLA profiles, (ii) infection of EBV, and (iii) traditional Cantonese diets, especially salted fish.<sup>7)</sup> The carcinogens in salted fish and other traditional Cantonese diet are still unknown and a systematic search for such substances is necessary. NPC arises commonly among Asian people but is quite rare in other parts of the world. It is suggested that undifferentiated NPC due to EBV infection may be associated with geographical and/or ethnic origin. On the other hand, in America, Chinese immigrants in the second and third generation seem to have lower risk of NPC than first generation Chinese.<sup>1)</sup> A possible explanation may be that EBV infections and environmental chemicals could act synergistically in the carcinogenic process.<sup>6)</sup> This hypothesis is supported by epidemiological and experimental evidences that viral replication and virally induced transformation were dramatically enhanced by chemical carcinogens. Therefore, chemical carcinogens could activate nasopharyngeal epithelial cells to EBV infection, or already carrying EBV, resulting in development of NPC.

## References

- 1) Burt RD, Vaughan TL, McKnight B: Descriptive epidemiology and survival analysis of nasopharyngeal carcinoma in the United States. *Int J. Cancer*; 52:549-556, 1992.
- 2) Butler AE, Colby TV, Weiss LM, Lombard C: Lymphoepithelioma-like carcinoma of the lung. *Am. J. Surg. Pathol.*, 13:632-639, 1989.
- 3) Chan JKC, Ng CS, Lau WH, Lo STH: Most nasal/nasopharyngeal lymphomas are peripheral T-cell neoplasms. *Am. J. Surg. Pathol.*, 11:418-429, 1987.
- 4) Dawson CW, Rickinson AB, Young LS: Epstein-Barr virus latent membrane protein inhibits human epithelial cell differentiation. *Nature*, 344:777-780, 1990.
- 5) Ferry JA, Sklar J, Zukerberg LR, Harris NL: Nasal lymphoma: A clinicopathologic study with immunophenotypic and genotypic analysis. *Am. J. Surg. Pathol.*, 15:268-279, 1991.
- 6) Ho JHC, Huahg DP, Fong YY: Salted fish and nasopharyngeal carcinoma in southern Chinese. *Lancet*, ii:626, 1978.
- 7) Krishnamurth S, Lanier AP, Dohan P, Lanier JF, Henle W: Salivary gland cancer in Alaskan natives, 1996-1980. *Hum. Pathol.*, 18:986-996, 1987.
- 8) Mills SE, Austin MB, Randall ME: Lymphoepithelioma-like carcinoma of the uterine cervix: a distinctive, undifferentiated carcinoma with inflammatory stroma. *Am. J. Surg. Pathol.*, 9:883-889, 1985.
- 9) Niedobitek G, Young LS, Sam CK, Brooks L, Prasad U, Rickinson AB: Expression of Epstein-Barr virus genes and of lymphocyte activation molecules in undifferentiated nasopharyngeal carcinoma. *Am. J. Pathol.*, 140:879-887, 1992.
- 10) Parshall DB, Stenstrom KW: Malignant lesions of the tonsil. *Radiology*, 60:564-571, 1953.
- 11) Saw D, Lau WH, Ho JHC, Chan JKC, Ng CS: Malignant lymphoepithelial lesion of the salivary gland. *Hum. Pathol.*, 7:914-923, 1986.
- 12) Senba M, Zhong XY: Immunohistochemical investigation of nasopharyngeal carcinoma using keratin, EMA, laminin, fibronectin, collagen type IV, laminin receptor, and laminin/collagen receptor antibodies. *Acta Med Nagasaki*, 38: (in press), 1993.
- 13) Senba M, Zhong XY: T-cell rich B-cell cross-reacting nasopharyngeal carcinoma. *Acta Med Nagasaki*, 38: (in press), 1993.
- 14) Senba M, Zhong XY: Proliferating cell nuclear antigen (PCNA), nuclear organizer regions (AgNORs), and mitotic indices in nasopharyngeal carcinoma. *Acta Med Nagasaki*, 38: (in press), 1993.
- 15) Shanmugaratnam K, Sobin LH: Histological typing of upper respiratory tract and ear: WHO international histological classification of tumors. 2nd Ed. Berlin, Springer-Verlag, 1991.
- 16) Shi SR, Goodman ML, Bhan AK, Pilch BZ, Sun TT: Immunohistochemical study of nasopharyngeal carcinoma using monoclonal keratin antibodies. *Am. J. Pathol.*, 117:53-63, 1984.
- 17) Shibata D, Tokunaga M, Uemura Y, Sato E, Tanaka S, Weiss LM: Association of Epstein-Barr virus with undifferentiated gastric carcinomas with intense lymphoid infiltration: Lymphoepithelioma-like carcinoma. *Am. J. Pathol.*, 139:469-474, 1991.
- 18) Swanson SA, Cooper PH, Mills SE, Wick MR: Lymphoepithelioma-like carcinoma of the skin. *Mod. Pathol.*, 1:359-365, 1988.
- 19) Wick MR, Scheithauer BW, weiland LH, Bernatz PE: Primary thymic carcinomas. *Am. J. Surg. Pathol.*, 6:613-630, 1985.
- 20) Zhang HY, Tao K, Shu HC, Glaser R: Expression of the Epstein-Barr virus genome in nasopharyngeal carcinoma epithelial tumor cell line. *Int. J. Cancer*, 46:944-946, 1990.
- 21) Zhong XY, Senba M, Gu JN, Itakura H: B-cell antigen marker expression on the nasopharyngeal carcinoma. *Zentralbl. Pathol.*, (in press), 1993.