Eur J Cancer Clin Oncol, Vol. 23, No. 9, pp. 1277–1281, 1987.
Printed in Great Britain

0277-5379/87$3.00+0.00

Nasopharyngeal Carcinoma in Israel: Epidemiology and Epstein–Barr Virus-Related Serology

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Abstract—*Epidemiological, histological and serological characteristics of nasopharyngeal carcinoma (NPC) were investigated. Included were 25 patients aged 10–70 with male to female ratio 2 : 1. Among 23 Jewish patients, 18 were of Asian–African (AA) and five of European (Eur) descent; two were Arabs (Ar). The dominant histological type among AA patients was undifferentiated carcinoma (UCNT) and among Eur squamous cell carcinoma (SCC).*

Elevated IgG and IgA antibodies to Epstein–Barr (EBV) viral capsid, early and nuclear antigens were observed in patients, as compared to 34 healthy controls matched by age, sex and ethnic origin. Although not statistically significant, antibodies to EBV were elevated in AA, as compared to Eur patients.

No significant differences in IgG and IgA antibodies to Herpes simplex, Cytomegalo and Varicella-zoster viruses were demonstrated among patients and controls.

The study suggests that NPC in Israel, as elsewhere, is associated with EBV and genetic or environmental factors may influence the prevalence of NPC among certain ethnic groups.

INTRODUCTION

THE ASSOCIATION of nasopharyngeal carcinoma (NPC) and Epstein–Barr virus (EBV) has been established world-wide by comparative serological studies in NPC patients [1–5]. NPC is the most prevalent ear, nose and throat tumor in male Chinese in south-east Asia [6, 7] and the fourth most common tumor in northern Africa [8]. The serological and clinical features of this tumor were found to be indistinguishable among different ethnic groups and different populations or geographic regions [9].

The present study was undertaken in an attempt to characterize the epidemiologial, histopathological, virological and clinical aspects of NPC in Israel, its relation to EBV and to other Herpes viruses.

MATERIALS AND METHODS

The study was done retrospectively and included NPC patients diagnosed between 1974 and 1982 in two major medical centers in the central coastal area of Israel. Information on therapy modalities was available for 19 patients. Four received radiotherapy alone, 14 received radiotherapy followed by chemotherapy. One patient received irradiation after excisional biopsy. Six patients died. Of those who survived, three had relapsed and were retreated and 14 are alive with no evidence of disease. Sera were obtained from patients, either at the time of diagnosis or during therapy. Twenty-five patients, who were followed from 3 months to 12 years, were included. Sequential sera were collected and stored at −20°C until assayed.

Thirty-four healthy subjects matched for age, sex and ethnic origin served as controls: nine patients had two matched controls each and the other 16 were matched to one control each.

Immunofluorescence tests: sera of patients and controls were tested simultaneously for the presence of IgA and IgG type antibodies to EBV viral capsid antigen (VCA) and EBV induced early antigen (EA-D type) and for IgG type antibodies to EBV nuclear antigen (EBNA) [10–12]. Determinations for the presence of IgG and IgA specific antibodies to Herpes simplex type 1 virus (HSV), Varicella-zoster virus (VZV) and Cytomegalo virus (CMV) were carried out by conventional indirect fluorescence assays. Fluorescein-conjugated rabbit anti-human IgG and goat-antihuman-IgA-fluorescein-

Accepted 23 January 1987.
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Table 1. Distribution of the study group by age, sex and ethnic origin

Age group (years)	Number of patients	Sex		Ethnic origin		
		Male	Female	AA	Eur	Ar
10–19	5	5	—	3	1	1
20–29	—	—	—	—	—	—
30–39	6	1	5	5	—	1
40–49	7	7	—	6	1	—
50–59	6	4	2	4	2	—
60–69	1	1	—	—	1	—
Total	25	18	7	18	5	2

isothiocyanate purchased from Behringwerke were employed. Sera were diluted two-fold starting at 1 :10 to 1 : 10,240. Geometric mean titers (GMT) were calculated and log titers were subjected to statistical analysis by Student's *t* test.

Histopathological classification: the relative frequency of specific histological types was determined according to the French scheme, which divides NPC into two subgroups: 1. squamous cell carcinoma (SCC); 2. undifferentiated carcinoma of nasopharyngeal type (UCNT), which includes all NPC other than SCC, undifferentiated, anaplastic, transitional cell and lymphoepithelial carcinoma [13, 14].

RESULTS

The group of patients consisted of 17 males and eight females; among them, 23 were Jews and two Arabs (Ar). The Jews were divided into two ethnic groups, according to country of origin of the father. Eighteen Sephardic Jews originating from the Mediterranean basin, Middle East or Yemen were designated Asian–African (AA) and five Ashkenazi Jews originating from western or eastern Europe were designated European (Eur).

The distribution of patients by age, sex and county of origin is presented in Table 1. Ages ranged from 12 to 62 years and were characterized by two peaks, an early one included five adolescent patients aged 11–14 years and a late one between 30 and 60 years old. Five out of seven females were 30–40 years old at the time of diagnosis. Among the Eur group there was a tendency for the disease to occur at a later age than in the AA group. Histologically, 18 cases were diagnosed as UCNT, including 10 lymphoepitheliomas, four undifferentiated carcinomas and four anaplastic carcinomas. All seven patients with SCC were categorized histologically as poorly or undifferentiated. Among the 18 AA patients, 16 were UCNT and two SCC, while among the Eur group, one patient had UCNT and four had SCC. The two Arabs had UCNT. At time of diagnosis, all 25 patients were classified as stage 1 or stage 2. The mean follow-up period for the two histological groups was not significantly different, death occurred among UCNT patients (6/18), whereas recurrences occured in both groups.

In 12 patients, the initial serum samples were obtained at the time of diagnosis. In the other 13, the first blood specimens were collected at the initial stage of therapy. The GMTs of EBV-related serological parameters (VCA and EA-IgA and IgG and EBNA) in the group tested before therapy did not differ from those tested during therapy ($P > 0.05$). Hence, all patients were considered as one group for comparison with the control group. Antibody levels to EBV-VCA and EA in both IgG and IgA class and IgG type antibodies to EBNA, are shown in Table 2.

Significant differences in all five assays between patients and controls were found ($P < 0.01$–0.001). Most patients had detectable IgA antibodies to VCA and to EA, whereas no such antibodies were present in any of the control subjects. There were no differences in IgG type VCA antibody titers between males and females (GMT 602.5 vs. 1000.0, respectively, $P > 0.05$) or between adolescents and adults (GMT 724.4 vs. 501.2, respectively, $P > 0.05$). During follow-up, which averaged 37 months (range 3–97 months), no consistent pattern of decrease in IgG and IgA type EBV antibodies was documented, although 14 patients were in complete remission.

Antibody response in the two histopathological groups is shown in Table 3. Differences were not significant for all antibodies tested between UCNT and SCC ($P > 0.05$).

Antibody levels in the two Jewish subgroups varied greatly, as shown in Table 4. The GMT of all types of EBV related antibodies were higher in the AA than in Eur group. However, differences did not reach statistical significance.

Antibody responses to HSV, VZV and CMV in both IgG and IgA classes of immunoglobulins are demonstrated in Table 5. No significant differences were found between patients and controls. Specific IgG antibodies to the three viruses were detected in the majority of the patients and controls, but specific IgA antibodies to these viruses were low or undetectable.

DISCUSSION

The present study investigated the epidemiologic, demographic, histopathologic and serologic profile of EBV and other Herpes viruses in NPC patients in Israel.

Although mainly an adult type tumor, we have found five adolescents among 25 patients with NPC, giving an incidence in this age group comparable to that found in Tunisia [15] and different from the U.S.A. [16].

Table 2. Distribution of antibody titers to EBV related antigens and GMT in NPC patients and in matched controls

Antibody type	Group	Serum dilution (reciprocal) < 10	10–40	80–320	⩾ 640	No. tested	GMT	*P*
VCA IgG	NPC	—	1	9	15	25	767.3	< 0.001
	Control	4	27	3	—	34	13.5	
VCA IgA	NPC	4	4	3	3	19	60.2	< 0.001
	Control	34	—	—	—	34	< 2.0	
EA IgG	NPC	4	11	6	3	24	34.6	< 0.01
	Control	32	2	—	—	34	2.2	
EA IgA	NPC	7	9	1	2	19	14.8	< 0.01
	Control	34	—	—	—	34	2.0	
EBNA IgG	NPC	1	16	2	—	19	25.1	< 0.01
	Control	14	3	—	—	17	6.3	

Table 3. Distribution and GMT of IgG and IgA-EBV specific antibody titers in NPC patients by histological type

Antibody type	Histological type	Serum dilution (reciprocal) < 10	10–40	80–320	⩾ 640	No. tested	GMT*
VCA IgG	SCC	—	—	4	3	7	501.1
	UCNT	—	1	4	11	16	812.8
VCA IgA	SCC	—	3	2	1	6	63.1
	UCNT	4	1	6	2	13	57.5
EA IgG	SCC	—	6	—	1	7	20.8
	UCNT	4	4	5	2	15	46.6
EA IgA	SCC	2	2	1	1	6	16.2
	UCNT	5	5	1	1	12	14.1
EBNA IgG	SCC	1	4	1	—	6	22.3
	UCNT	—	12	2	—	14	33.8

*Differences in GMT in each category are not significant ($P > 0.05$).

*Table 4. EBV antibody titers (GMT) in Jewish patients and matched controls by country of origin**

	VCA IgG		VCA IgA		EA IgG		EA IgA	
	Eur	AA	Eur	AA	Eur	AA	Eur	AA
Patients	320.0	812.8	25.1	95.4	17.3	47.8	6.3	21.3
Controls	23.4	15.8	<2.0	<2.0	<2.0	6.3	<2.0	<2.0

*Number of patients by country of origin: AA, 18; Eur, 5. Number of controls by country of origin: AA, 22; Eur, 10.
Differences in GMT between patients and controls are significant ($P < 0.01$–0.0001). Differences in GMT by country of origin are not significant ($P > 0.05$).

Histologically, the diagnosis of poorly or undifferentiated SCC in 28% of NPC patients in our study group is close to the incidence of this tumor found in several of the studies reported from north America or Singapore [9]. Of interest is the ethnic distribution of NPC in the Jewish population. A large proportion of the Jewish population in Israel are first generation, of which about 60% come from Asian–African countries and 18 of the Jewish patients belong in this group. Moreover, 16 (among them three adolescents) out of the Asian–African group and the two Arab patients had UCNT.

*Table 5. Antibody response to HSV, CMV and VZV in NPC patients and in matched controls**

Virus	Antibody type	Patients No. tested	Patients MGT	Controls No. tested	Controls MGT
HSV	IgG	21	269.1	29	204.1
	IgA	20	1.4	22	1.6
CMV	IgG	18	57.5	28	16.2
	IgA	15	1.7	21	1.1
VZV	IgG	21	5.7	28	7.1
	IgA	18	1.0	22	1.0

*Differences in GMT between patients and controls not significant.

The higher incidence of NPC among Jews of Asian–African descent does not seem to be incidental. It conforms with the cumulative data of the Israel Cancer Registry for the years 1972–1976 [17], which reported 81 cases of NPC, of whom 55 were of Asian–African descent and 26 were European (2 : 1). This distribution of NPC in the two ethnic subgroups and the dissociation of the histological types between them may be related to genetic or to environmental factors. Indeed, previous data indicated that both factors are associated with increased incidence of NPC in certain populations [7, 18–20]. HLA typing was not feasible due to the retrospective nature of our study.

The serological findings conform with previous reports in two major points. First, antibodies to different EBV antigens were present in patients in higher titers than in matched controls. Secondly, the most specific EBV antibody response was of the IgA type [10, 12, 21].

The specificity of the IgA type antibody response to EBV and the usefulness of determination of this class of antibodies was demonstrated in mass surveys in endemic areas [20]. It was considered a sensitive marker for clinical and prognostic evaluation; long term survival was correlated with initial lower levels and with a gradual loss of these antibodies [22–24]. Levels of IgA type EBV antibodies were determined in a normal population and found to be significantly lower than in patients with NPC [16]. In our study, almost no such antibodies were detected in the control subjects. It has also been observed by others that IgA type antibodies were frequent and in higher levels in UCNT whereas in SCC type tumors the titers resembled the control groups and high IgA antibodies correlated with biopsy-proven EBV-DNA or EBNA [9, 10, 13]. We did not observe significant differences in EBV antibody profile between the two histologcal groups. This may be attributed to the small number of patients or more likely to the fact that all the SCC were of the poorly or undifferentiated type. This tumor, which is considered a more advanced carcinomatous tumor than the well differentiated type was shown to be associated with EBV similarly to UCNT [9]. Both the frequency and the level of EBNA antibodies in patients were also increased compared to the general population, but no differences were detected in relation to the histological types, unlike the finding among Chinese patients [9]. In addition, we were unable to identify long-term survivors based on the pattern of their antibody response.

We previously noted that Hodgkin's patients of Asian–African origin mounted a higher EBV antibody response, as compared to patients of European descent, while no such differences were observed in a healthy control population [25, 26]. Similarly, in the present study, there were no differences in EBV antibody levels among healthy control subjects of the two Jewish subgroups, while a difference seemed to be demonstrated among patients, although it did not reach statistical significance.

Finally, we looked at antibody response to other Herpes viruses and unlike other investigators [27] we found no differences in either IgG or IgA type antibody level to HSV, VZV or CMV between patients and controls substantiating the observation that IgA antibodies to EBV antigens are specific in NPC.

It seems of interest to further investigate possible contribution of genetic and environmental factors to the development of EBV associated NPC.

Acknowledgements—The authors thank Dr. S. Leventon-Kriss and Mr. Y. Sayar for tests of antibodies to HSV, VZV and CMV.

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