Associations between B-cell non-Hodgkin lymphoma and exposure, persistence and immune response to hepatitis B

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Supplemental Methods

Study design

This is a hospital and community-based case-control study in Israel and the West Bank including Palestinians and Israelis (Supplemental Methods). For purposes of analysis Israeli citizens of Muslim origin were grouped together with Palestinians.

Study population

Inclusion criteria

We enrolled incident cases (target: within 18 months of diagnosis), aged ≥18 years, with pathologically confirmed CD20+ (or other B-cell marker+) B-NHL, and healthy controls aged ≥18 years accompanying or visiting out- and inpatients[1] in participating centers.

Exclusion criteria were inability to provide written informed consent; HIV positivity; being a blood relative of a case; and being a spouse of an enrolled case for Palestinian controls (given frequent cousin marriages).

Recruitment:

Cases were recruited in: a) Hadassah–Hebrew University Medical Center (HMC), a tertiary center on two campuses between October 2010 - March 2014 (N=507); b) Chaim Sheba, Meir, Rambam and HMC Medical Centers, university hospitals in the center and north of Israel, which participated in an uncompleted case-control study conducted in 2003 (Epilymph-Israel) (N=86) ; c) Augusta Victoria Hospital (AVH) in East Jerusalem, National Hospital of Nablus (NHN) and Al-Husein Hospital (AHH) in Beit Jalla in the West Bank (N=170), 2009-2013 and, d) using the West Bank Cancer Registry files, 2009-2013 (N=60).

Controls were recruited in: a) HMC for the Israeli cases (N=414); b) AVH, NHN, AHH, and 16 ambulatory health centers in the West Bank for the Palestinian cases (N=394).

Study variables

Participants were interviewed in Hebrew, Arabic, Russian or English utilizing a questionnaire adapted from that used in the European Epilymph study[2]. Questionnaire items included demographic characteristics as well as family and medical history (reported separately).

Hepatitis B Biomarkers

Serum samples were obtained and tested for the presence of HBsAg, anti-HBc, anti-HBs, hepatitis B e antibodies (anti-HBe) and hepatitis B e antigen (HBeAg), by ELISA (Roche Elecsys®, Basel, Switzerland) at AVH.

Anti-HBc+ individuals were considered "exposed" to HBV. Among these, individuals who were HBsAg+ were considered to have persistent infection while those HBsAg- were considered to have occult infection (OBI). Individuals who demonstrated anti-HBs+ with anti-HBc- were considered immune via vaccine, while anti-HBs+ with anti-HBc+ were considered naturally immune via exposure, although some of the individuals designated "vaccinated" might actually be naturally immune. Individuals who were anti-HBc+ and anti-HBs- were considered to lack immune response following viral exposure.

Statistical analysis

Distribution of baseline variables in cases and controls were assessed using two-sided Fisher's exact and χ^2 tests. Conditional logistic regression models were built to test the association [reported as ORs and 95% confidence intervals (CIs)] between B-NHL status (and subtypes DLBCL and FL) and hepatitis biomarkers- stratified by population (Israelis/Palestinians), sex and age categories (four year groupings); and adjusted for factors associated with the outcome (B-NHL) or exposure (hepatitis serology) including marital status, education (yrs), family history of hematopoietic malignancies in first-degree relatives. Individuals with missing data for the exposure variable of interest were excluded. Heterogeneity was assessed between population groups and serologic markers by adding an interaction term into the conditional logistic model.

Agreement of pretreatment serology results performed for clinical purposes and post-treatment within the study among a subgroup of cases was assessed using Kappa scores with 95% CIs.

Based on an assumed average prevalence (Israelis and Palestinians) in controls of 2% for HBV, two sided test with α=0.05; a study with 800 cases and 800 controls provided at least 85% power to detect an OR of 2.5.

Sensitivity analyses were performed for the OR estimations, a) using different stratification strategies to assess the stability of the results, b) for overall B-NHL and DLBCL associations excluding spouse controls, c) excluding those with borderline anti-HBc results, and d) interpreting borderline results as negative or positive.

Ethics

The study was approved by the Research Ethics Committee (IRB) of the Hadassah University Hospital and other participating institutes in Israel and the Institutional Review Board of the Palestinian Ministry of Health. All participants provided written informed consent.

Role of the funding source

The funding sources played no role in the design, execution or analysis of the study.

Supplemental Table 1

Supplemental Table 1. Demographic characteristics, medical and family history for cases and controls, overall B-NHL, DLBCL and FL

Characteristics		DLBCL		FL		Overall B-NHL		Controls		P*
		N	(%)	N	(%)	N	(%)	N	(%)	
Total No.		427	(40.7)	186	(27.7)	823	(100)	808	(100)	
Population	Israeli	210	(49.2)	143	(76.9)	516	(62.7)	414	(51.2)	<0.001
	Palestinian	217	(50.8)	43	(23.1)	307	(37.3)	394	(48.8)	
Religion	Jewish	204	(48.1)	139	(76.0)	499	(61.6)	405	(50.4)	<0.001
	Muslim	200	(47.2)	38	(20.8)	282	(34.8)	369	(45.9)	
	Christian	16	(3.8)	6	(3.3)	25	(3.1)	24	(3.0)	
	Other	4	(0.9)	0	(0.0)	4	(0.5)	6	(0.7)	
Sex	Male	200	(46.8)	97	(52.2)	413	(50.2)	352	(43.6)	<0.01
	Female	227	(53.2)	89	(47.8)	410	(49.8)	456	(56.4)	
Age	<34	74	(17.3)	12	(6.5)	97	(11.8)	99	(12.3)	<0.01
(years)	35-54	132	(30.9)	59	(31.7)	253	(30.7)	285	(35.3)	
	55-64	83	(19.5)	59	(31.7)	199	(24.2)	196	(24.2)	
	65-74	82	(19.2)	30	(16.1)	158	(19.2)	163	(20.2)	
	≥75	56	(13.1)	26	(14.0)	116	(14.1)	65	(8.0)	
Marital status	Single	46	(10.9)	10	(5.4)	70	(8.5)	53	(6.6)	<0.01
	Married	305	(71.9)	148	(79.6)	606	(73.9)	715	(88.5)	
	Other	73	(17.2)	28	(15.0)	144	(17.6)	40	(4.9)	
Education†	0-8	141	(33.6)	32	(17.3)	206	(25.3)	178	(22.2)	0.14†
(years)	9-12	115	(27.4)	56	(30.3)	235	(28.9)	218	(27.3)	
	>12	164	(39.0)	97	(52.4)	372	(45.8)	404	(50.5)	
First-degree relatives with hematopoietic malignancy	Yes	42	(10.2)	15	(8.6)	83	(10.7)	55	(6.8)	<0.01

B-NHL=B-cell non-Hodgkin lymphoma; DLBCL= diffuse large B-cell lymphoma; FL=follicular lymphoma; P* calculated for overall B-NHL versus controls; Values were missing for < 5% for exposure variable; †Education was not significantly associated with case control status but highly associated with HBV exposure, hence it was included in multivariable models.

References

1. Asomaning K, Miller DP, Liu G, Wain JC, Lynch TJ, Su L, et al. Second hand smoke, age of exposure and lung cancer risk. Lung cancer. 2008;61(1):13–20.

2. Besson H, Brennan P, Becker N, Nieters A, De Sanjosé S, Font R, et al. Tobacco smoking, alcohol drinking and non-Hodgkin's lymphoma: A European multicenter case-control study (Epilymph). Int J Cancer. 2006;119(4):901–8.