# Hodgkin's Lymphoma in the Bedouin of Southern Israel: Epidemiological and Clinical Features

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## Abstract

**Background:** A previous study on Hodgkin's lymphoma in southern Israel found that Bedouin patients had an increased rate of Epstein-Barr virus expression in their tumor cells.

**Objectives:** To determine the influence of the patients' communities on the pattern of disease in HL.

**Methods:** We compared the clinical features, demographic data, stage at diagnosis, treatment modality and outcome, as well as laboratory findings, in four community-based subgroups. These groups comprised kibbutz residents (n=11), Bedouin (n=19), new immigrants from the former USSR (n=22), and town-dwellers (n=82).

**Results:** The Bedouin patients differed significantly from the new immigrants and town-dwellers, particularly regarding the rate of EBV sequences in the tumor tissues, and a poorer response to treatment. The kibbutz patients did not differ significantly from the other populations regarding most of the parameters studied, but showed an intermediate expression of EBV antigens compared to Bedouin patients and the rest of the cohort.

**Conclusions:** This study indicates that HL may behave differently in different population groups in a given geographic area. Notably, the Bedouin patients showed markedly different clinical and biological patterns of this malignancy.

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In a previous study of Hodgkin's lymphoma in southern Israel we found a relatively low association of the tumor with Epstein-Barr virus sequences (30%), except among Bedouin patients who exhibited a significantly increased rate of EBV expression [1]. We propose that the lifestyle of the communities to which our patients belonged may influence the various clinical and biological features of the disease. Many of the Bedouin preserve their traditional mode of living and thus differ from other inhabitants of southern Israel. We also suggest that the kibbutz, with its high standard of health care and high level of physical contact between its children, may have an impact on the pattern of HL that is not seen in the other population groups. A third group of HL patients – new immigrants to Israel from the former USSR – was also included due to the apparent high rate of malignancies, including HL, in this population. The fourth group, comprising town-dwellers, served as the reference group in spite of its heterogeneity.

By carrying out a comparative study of our cohort of patients from these four types of population, we hope to be able to contribute to an understanding of the epidemiology of HL, perhaps in the context of the "three diseases" postulate [2,3]. This thesis supports the heterogeneous nature of HL. Thus, the patterns of the disease vary in different age and socioeconomic groups, mainly in relation to the incidence of EBV infection.

## Methods

Our previous study was expanded to include 134 HL patients. The clinical features included gender, age groups (0–9, 15–34, and 50 or more years, as described by Armstrong [3]), the histological subtype (according to the REAL classification [4]), the stage of the disease at diagnosis (modified Ann Arbor classification: I-IIA versus IIB-IVB [5,6]), and the presence of bulky disease. Treatment modalities included radiotherapy and/or chemotherapy (usually MOPP and/or ABVD). The outcome was grouped as follows: no evidence of disease at last visit, alive with disease or dead of disease, and dead due to causes other than HL. The presence of EBV sequences was studied immunohistochemically with the LMP antibody (Dako, Denmark) and confirmed in 64 cases by EBER-RNA *in situ* hybridization, as described [1]. We also studied the expression of p53, bcl-2, mdm-2 and CD3O by immunohistochemistry. An apoptotic index was calculated using the TUNEL assay [ApopTag, the apoptotsis detection kit (Oncor, Gaithesburgh, USA)] as described previously [7].

HL = Hodgkin's lymphoma
EBV = Epstein-Barr virus

The origin of each patient (kibbutz, Bedouin and town-dwellers) was identified by the residential address at diagnosis. New immigrants who arrived in Israel from the former USSR after 1990 were identified from the hospital records.

For contingency table analysis, the Chi-square or Fisher's exact tests were used as appropriate. Analysis of continuous variables was performed using one way ANOVA. A multiple logistic regression analysis was used to estimate the adjusted risk factors for an unfavorable outcome (including patients who were either dead of disease or alive with disease) as compared with patients who were alive without evidence of disease, at last follow-up.

## Results

Of the 134 patients, 58 were females and 76 were males; 5 were aged 4–9 years, 63 were aged 15–34, and 28 were 50 years or older. HL was typed as lymphocyte predominant subtype in 6 cases, mixed cellularity in 38, nodular sclerosis in 80, and lymphocyte depleted HL in 6 cases. Diagnosis was made at an early stage (I-IIA) in 40 patients (39.6%) and at an advanced stage in 61 (60.4%). Bulky disease was present in 22 patients (20.9%). Radiotherapy was administered to 38 patients (40.4%) and MOPP/ABVD to 72 (83.7%). Eighty-three patients (69.2%) had no evidence of disease at their last visit, and 37 (30.8%) had died of the disease or were alive with disease.

Table 1 compares the findings among the Bedouin and in all other groups. The Bedouin patients were younger and included significantly more males (78.9% vs. 53.0%, $P=0.03$). More Bedouin presented with mixed cellularity HL ($P=0.16$) and at more advanced stages (85.7%, $P=0.04$); 58.3% received ABVD and/or MOPP, as compared with 87.8% in the rest of the population ($P=0.02$), and 47.4% showed resistance to treatment (alive with disease or dead of disease) as compared with 27.7% in the rest of the cohort ($P=0.09$). When compared with town-dwellers, the Bedouin patients showed an even gloomier outcome (no evidence of disease in only 52.6% in the Bedouin as compared with 75.7%, $P=0.049$). Patients without evidence of tumor at the last visit (no evidence of disease) were followed for a mean period of 61.9±46.3 months as compared to 38.7±37.9 months for those patients who fared worse (alive with disease or dead of disease).

We compared the laboratory findings between the Bedouin and the other population groups (data not shown). EBV sequences were significantly more frequent [Table 2] and mdm-2 less frequent in the Bedouin. Comparison of EBV expression in the town-dwellers and in the immigrants with that of the Bedouin patients showed a statistically significant difference ($P=0.02$ and $P=0.05$ for the town-dwellers and the immigrants, respectively).

Among Jewish patients, kibbutz residents exhibited a positive EBV expression more frequently (36.4%). Bedouin patients had a significantly higher apoptotic index (17,

**Table 1.** Hodgkin lymphoma: comparison of demographic and clinical features in Bedouin versus other patients

| Demographic and clinical features | Bedouin No. (%) | Other groups No. (%) | *P* value |
|---|---|---|---|
| Gender | | | |
| Female | 4 (21.1) | 54 (47.0) | |
| Male | 15 (78.9) | 61 (53.0) | 0.03 |
| Age (yr) | | | |
| 15–34 | 8 (80.0) | 55 (67.9) | |
| >50 | 2 (20.0) | 26 (32.1) | 0.35 |
| Type | | | |
| Mixed cellularity | 7 (46.7) | 31 (30.1) | |
| Nodular sclerosis | 8 (53.3) | 72 (69.9) | 0.16 |
| Stage | | | |
| I-IIA | 2 (14.3) | 38 (43.7) | |
| IIB-IVB | 12 (85.7) | 49 (56.3) | 0.04 |
| Bulky | | | |
| Yes | 3 (25.0) | 19 (20.4) | |
| No | 9 (75.0) | 74 (79.6) | 0.48 |
| Outcome | | | |
| No evidence of disease | 10 (52.6) | 73 (72.3) | |
| Alive with disease + dead of disease | 9 (47.4) | 28 (27.7) | 0.09 |
| Radiotherapy | | | |
| Yes | 2 (18.2) | 36 (43.4) | |
| No | 9 (81.8) | 47 (56.6) | 0.10 |
| Chemotherapy | | | |
| ABVD/MOPP | 7 (58.3) | 65 (87.8) | |
| Other | 5 (41.7) | 9 (12.2) | 0.02 |

**Table 2.** EBV sequences in Hodgkin lymphoma by population group

| Type of population | LMP positive (%) | LMP negative (%) | *P* value* |
|---|---|---|---|
| Kibbutz | 4 (36.4) | 7 (63.6) | 0.34 |
| Bedouin | 10 (52.6) | 9 (47.4) | – |
| Immigrants | 5 (22.7) | 17 (77.3) | 0.05 |
| Town-dwellers | 21 (25.6) | 61 (74.4) | 0.02 |

* The LMP (of EBV) expression is compared for each population group (kibbutz, immigrants and town-dwellers) with the Bedouin group.

**Table 3.** Multivariable logistic regression of the association of prognostic factors and demographic variables with outcome* in 90 patients diagnosed with HL

| Variable | Odds ratio** | 95% confidence interval | *P* value |
|---|---|---|---|
| Type of population | | | |
| Bedouin (compared to Jewish) | 8.7 | 1.6–48.0 | 0.01 |
| Stage | | | |
| IIB-IVB (compared to I-IIA) | 9.3 | 2.1–42.4 | 0.004 |
| LMP*** | | | |
| Positive (compared to negative) | 6.3 | 1.3–31.2 | 0.02 |

* Outcome, including patients who are either dead of disease or alive with disease as compared with patients who are alive without evidence of disease at last follow-up.

** Odds ratios are adjusted for age and gender.

*** LMP for IHC expression of EBV

89.5%); namely, above the median value as compared with 71 (62.1%) for the rest of the population, $P=0.02$]

A multivariable logistic regression analysis was performed to identify adjusted risk factors for an unfavorable outcome (including patients who were either dead of disease or alive with disease) as compared with patients who were alive without evidence of disease at the last follow-up. Table 3 shows that Bedouin have a higher risk of developing resistant disease than Jewish patients. This risk was 8.7-fold ($P=0.01$) when adjusted for age, gender and other factors. Advanced stage and positive EBV expression were statistically significant risk factors as well (odds ratio 9.3 and 6.3 for stage IIB-IVB and positive LMP, respectively).

## Discussion

In our investigation of the impact of the type of community to which our patients belonged on the clinical and biological features of HL in southern Israel, the results support our previous findings that Bedouin patients with HL stand out as a separate group regarding several features of this malignancy [1].

The proportion of male patients is significantly higher among the Bedouin, and more of the Bedouin patients are diagnosed at an earlier age. They present at more advanced Ann Arbor stages, and the rate of EBV-related sequences in their tumor cells is much higher than in any of the other groups. Bedouin HL patients show an unfavorable response to treatment. This finding may be due to a very poor compliance among this population group (S. Ariad, personal communication), which was much more marked in male patients who predominate in this population group. The inadequate treatment is demonstrated by the finding that the Bedouin received less radiotherapy and significantly less chemotherapy with ABVD and/or MOPP. The finding of a higher stage at diagnosis as well as an increased apoptotic index, shown by us to be associated with a worse prognosis [7], may indicate that the poor outcome is not entirely due to a lack of compliance. In fact, the origin of the patients in the Bedouin community by itself was an independent unfavorable risk factor. This was true also for a positive expression of EBV-related antigens. This finding lends support to the conclusions by Enbald et al. [8] and others [9] in 1999 regarding the role of EBV expression as a prognostic factor in HL.

The multivariable analysis included 90 patients (67% of the cohort) due to missing data. Comparison of disease outcome between patients included and those not included in the analysis showed no differences (an unfavorable outcome in 30.8% and 31.3%, respectively; $P=1.0$).

On the other hand, the Bedouin patients presented with a comparable level of p53 expression in the Hodgkin-Reed-Sternberg cells in their biopsies but significantly less mdm-2 and also, to some extent, less bcl-2 in the tumor cells. Brink et al. [10] have shown that low p53 and high bcl-2 expression in Hodgkin-Reed-Sternberg cells predict a poor clinical outcome in HL. Our finding of high p53 expression with a mdm-2 negative phenotype may be predictive of p53 gene mutation in these cells. This has yet to be confirmed [11].

We expected that kibbutz patients would show features of the disease different from those of town-dwellers or new immigrants from the former USSR, but the features of HL in the kibbutz patients did not differ from those of HL in the other population groups.

The diversity shown in the clinical and biological characteristics of HL in our cohort of patients reflects that found in countries with different socioeconomic conditions. Bedouin patients, with their high level of EBV-associated sequences, their higher proportion of the mixed cellularity histological subtype and their poorer response to therapy, would represent the features of HL in a developing country [2,3].

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