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Anti-LAV/HTLV-III Antibodies in High and Low Risk Groups[1]

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Introduction

Serum antibodies to LAV/HTLV-III (lymphadenopathy-associated virus/human T-lymphotropic virus type III) [1–3] have been found in the majority of patients with AIDS (acquired immunodeficiency syndrome) and in AIDS-related syndromes as well as in groups of individuals at increased risk for AIDS such as intravenous drug users, hemophiliacs, homosexual/bisexual men and in partners of patients with AIDS [4–7]. Anti-LAV/HTLV-III antibodies have also been detected in a minor but not negligible proportion of subjects at no known risk for AIDS. Of these latter individuals, antibody-positive donors represent a group of particular epidemiological importance, since they can transmit LAV/HTLV-III infection to blood recipients through blood transfusion.

Here we report an update of anti-LAV/HTLV-III antibody prevalence among Italian individuals at low and high risk of acquiring LAV/HTLV-III infection. Next to the prevalence of antibodies, we have studied, on stored samples, at what time LAV/HTLV-III infection had first occurred and how the percentage of seropositivity had increased over the years.

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Material and Methods

Subjects

High Risk Groups. A total of 1,107 intravenous drug users, 115 hemophiliacs and 94 homosexual/bisexual men were tested for anti-LAV/HTLV-III antibody. Among drug addicts, 602 individuals were either outpatients on methadone maintenance or inpatients on infectious diseases wards in hospitals for acute viral hepatitis (group A); sera were collected from 144 of these individuals from March 1985 to March 1986 and from 458 subjects during the years 1979–1984. The remaining 505 drug users (group B) had voluntarily entered a therapeutic community during the years 1979 to March 1986 and had no parenteral drug exposure or sexual contacts while they were staying in the community; sera were collected from March 1985 to March 1986. In addition, 115 asymptomatic hemophiliacs treated in the last 5 years with commercial concentrates from USA-derivated plasma and 94 asymptomatic homosexual or bisexual men were also examined.

Low Risk Group. From June to November 1985, a total of 48,266 volunteer blood donors attending 39 different Blood Transfusion Centers of Lombardy were screened for anti-LAV/HTLV-III antibody. Individuals examined (75% males, 25% females, mean age 35 years, range 18–65 years) roughly represent one-fifth of the donors of Lombardy. Among them, 43,163 were regular donors while 5,103 were at their first donation (occasional donors).

Methods

All serologic tests were performed by enzyme immunoassay (EIA) using commercially available kits (Abbott Laboratories, USA; Pasteur, France; Organon-Teknika, The Netherlands; Ortho-Diagnostic Systems, USA; Sorin Biomedica, Italy). Samples were considered positive when they were repeatedly reactive by EIA. All EIA-reactive samples collected from blood donors were retested by Western Blot (WB) [8], using our own reagents or performed strips (BioRad, USA).

Results

High Risk Groups

As shown in table I, among intravenous drug addicts, the overall anti-LAV/HTLV-III prevalence in sera collected from March 1985 to March 1986 was 49.3% in group A and 40.3% in group B. Testing of stored samples collected since 1978 from drug users of group A revealed that LAV/HTLV-III infection had first occurred starting from 1981 with a significant increase of infections in successive years (fig. 1). As concerns group B, anti-LAV/HTLV-III was again absent in drug addicts admitted in the therapeutic community before 1981, while the percentage of antibody seropositivity rose progressively up to 53% in individuals admitted in 1985 to March 1986 (fig. 1). Noteworthy is that all individuals who were anti-LAV/HTLV-III nega-

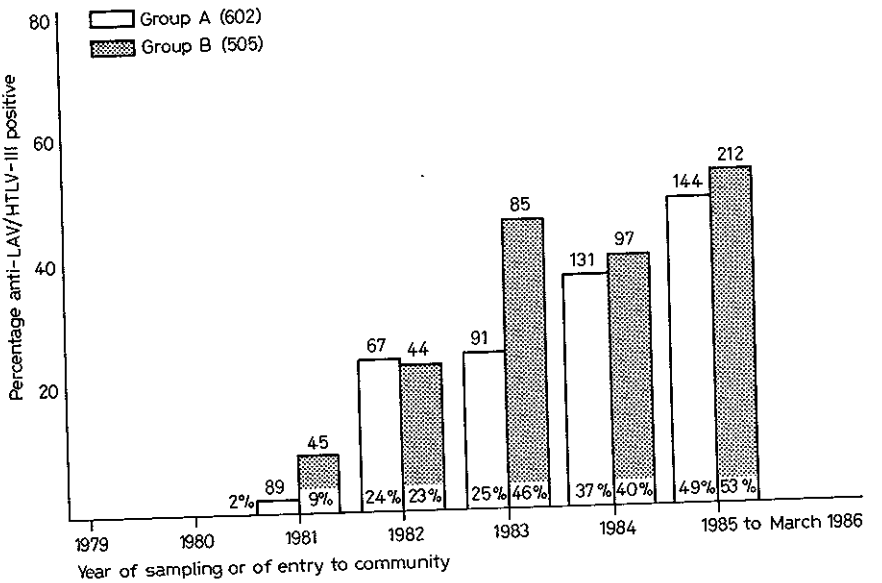

Fig. 1. Percentage of anti-LAV/HTLV-III in two groups of intravenous drug users by year of sampling (group A) or by year of admission to a therapeutic community (group B).

tive when admitted to the community, remained negative during their stay (1–6 years).

The prevalence of anti-LAV/HTLV-III antibody among hemophiliacs treated during the 1980–1985 time interval with commercial concentrates was 40.8% (table I). No significant difference was found between antibody presence and type of concentrates used (factor VIII or IX). Antibody seropositivity was found to correlate with the concentrates consumption, since anti-LAV/HTLV-III was present in 100% of hemophiliacs treated with more than 40,000 IU (median annual dose) and in 8% of those treated with less than 5,000 IU.

Finally, 11 (11.7%) of 94 homosexual/bisexual men were found to be anti-LAV/HTLV-III positive (table I). No correlation between the presence of anti-LAV/HTLV-III and markers of hepatitis B and hepatitis delta infections was seen.

Table I. Prevalence of anti-LAV/HTLV-III antibody in individuals at increased risk of infection (sera collected from March 1985 to March 1986)

Group	n	Anti-LAV/HTLV-III positive	
		n	%
Intravenous drug users			
Group A	144	71	49.3
Group B	505	204	40.3
Hemophiliacs	115	47	40.8
Homosexual/bisexual men	94	11	11.7

Table II. Anti-LAV/HTLV-III in Italian blood donors

Blood donors		EIA positive		WB positive					
		cases		cases		sex		age, years	
		n	%	n	%	M	F	mean	range
Regular	43,163	13	0.03	4	0.009	4	–	34	25–40
Occasional	5,103	15	0.29	9	0.176	7	2	25.5	22–33
Total	48,266	28	0.058	13	0.027	11	2	28.3	22–40

Low Risk Group: Blood Donors

As is reported in table II, 28 (0.058%) blood donors were found to be repeatedly reactive by EIA. Of these, 13 were confirmed positive, whereas 15 were found to be negative when tested by WB. Immunoprecipitation bands were observed at 18, 25, 41 and 65 kilodaltons in all donors except 2 who showed p25 and gp41 bands. Sample/cut-off absorbance ratios were found to be much lower (median value 1.8, range 1.03–10.2) among donors EIA positive but WB negative than in those found positive by both EIA and WB (median value 10.4, range 2.2–16.5). More in detail (fig. 2), 10 (66.6%) of 15 donor specimens not confirmed by WB exhibited sample/cut-off ratios less than 2, whereas 8 (61.5%) of 13 WB-positive specimens showed ratios higher than 10. Noteworthy is that two EIA weak positive samples (sample/cut-off ratios respectively of 2.2 and 2.4) were confirmed 'true'-positive by WB.

Of the 13 donors positive by both EIA and WB (table II), 4 (all males, mean age 34 years, range 25–40 years) were regular donors, while 9 (7 males, 2 females, mean age 25.5 years, range 22–33 years) were occasional donors.

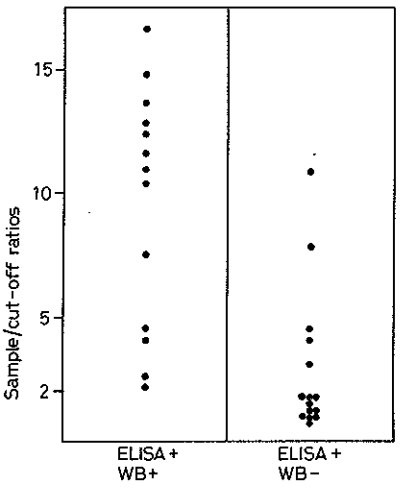

Fig. 2. Sample/cut-off EIA absorbance ratios in blood donors found anti-LAV/HTLV-III positive or negative by WB.

Therefore, the final anti-LAV/HTLV-III prevalence was 0.027%, with a significant difference between regular and occasional donors (0.009 vs. 0.176%, $p<0.01$). At least 11 (85%) of 13 anti-LAV/HTLV-III 'true'-positive donors were individuals at increased risk: 7 subjects (5 males and 2 females) had a past history of intravenous drug use; 3 were bisexuals; 1 was a sexual partner of an anti-LAV/HTLV-III-positive female intravenous drug user; 1 donor refused to give information, while the last 1 (regular donor) apparently had no known risk factors. When interviewed, antibody-positive regular donors did not believe they were high risk.

Discussion

The discovery of LAV/HTLV-III was instrumental in the development of several in vitro diagnostic techniques for the detection of specific antiviral antibodies. Availability of such methods which include EIA, indirect immunofluorescence assay, radioimmunoprecipitation and WB, has proved essential in order to study the epidemiologic pattern of LAV/HTLV-III infection and to prevent the spread of the virus, through blood transfusion, by massive serological screening of the blood donors.

Several studies performed in the USA and in Europe have shown that anti-LAV/HTLV-III antibody is very frequent in some groups of individuals (high risk groups) such as promiscuous homosexual/bisexual men, intravenous drug users, hemophiliacs treated with unheated concentrates, sexual partners of patients with AIDS and babies born to LAV/HTLV-III carrier mothers. On the other hand, in the USA anti-LAV/HTLV-III has also been detected in less than 1% of blood donors.

From our data as well as from those obtained from other studies in Italy [5, 7, 9], the following points emerge: (1) Parenteral drug users are indeed at very high risk for acquiring LAV/HTLV-III infection. Among them, infection was absent before 1981, while a progressive increasing spread during the successive years was observed. (2) Anti-LAV/HTLV-III prevalence in Italian homosexual/bisexual males was lower than that reported in the USA [4]. This may be due to the different level of sexual promiscuity, generally believed to be lower for Italian than for US homosexual/bisexual men. (3) A high percentage of hemophiliacs was infected by LAV/HTLV-III in recent years. This fact was quite expected since they were treated exclusively with concentrates made in the USA and therefore were exposed to the same high risk of infection of US hemophiliacs. (4) The risk of transfusion-associated AIDS in our country seems at present to be an alarming but relatively small problem. Self-exclusion from blood donation of individuals at high risk for LAV/HTLV-III infection combined with implementation of careful blood donor screening for AIDS risk factors and for LAV/HTLV-III antibody are the measures to be applied for ensuring the safety of blood supply. (5) The EIA test for the screening of blood donors should be the most sensitive available in order to avoid false-negative results. The possible loss of specificity, as a consequence of maintaining high sensitivity, needs the confirmation of all presumptive positive reactions by the use of a test with greater specificity, such as WB.

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