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Human T-Lymphotropic Virus Types 1 and 2 Are Rare Among Persons who inject drugs in Eastern Europe

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Abstract

Background—In Europe, human T-lymphotropic virus (HTLV) type 2 mainly occurs among persons who inject drugs (PWID) with prevalence up to 15% and HTLV-1 among general population with prevalence less than 1%. However, there is no data regarding the prevalence of HTLV-1 or HTLV-2 in Eastern European PWID population where HIV prevalence is relatively high. We aimed to determine the prevalence and genotypes of HTLV-1/HTLV-2 among PWID and healthy volunteers in Estonia.

Methods—The study included 345 PWID and 138 healthy volunteers. The presence of HTLV-1/HTLV-2 was determined by nested PCR; positive and negative controls were used in every PCR run.

Results—The analysed PWID resembled the PWID of HIV epidemic in Estonia: mainly male (79%) with median age of 30 years (interquartile range [IQR] 25–34), and prolonged duration of intravenous drug usage (11 years; IQR 7–14). The prevalence exposure to blood-borne viral infections was high – 50% were HIV positive, 88% hepatitis C positive, 67% hepatitis B positive. Of PWID, 64% reported receptive needle sharing in the past and 18% at least once a month during last six months. None of the PWID carried HTLV-1 but there was a case of HTLV-2 (prevalence 0.3 %; 95% CI 0.1–1.6). All healthy volunteers were HTLV-1 and HTLV-2 PCR negative.

Conclusion—This is the first study investigating the prevalence of HTLV-1/HTLV-2 among high risk population and healthy volunteers in Eastern European region. Our results suggest that despite other widely spread blood-borne infections (e.g. HIV, HBV, HCV) HTLV-1 and HTLV-2 are rare among PWID in Estonia.

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**The institution where the work was conducted

Keywords

HTLV-1/2; HIV; co-infection; blood-borne infection

Introduction

Human T-lymphotropic virus (HTLV) is a retrovirus with four distinct types of which HTLV-1 and HTLV-2 are the most prevalent. While their transmission routes are the same (transfusion of contaminated blood, mother to child, sexual contact), the clinical outcome of these viruses is quite different. HTLV-1 causes adult T-cell leukaemia/lymphoma or HTLV-1 associated myelopathy/tropical spastic paraparesis in up to 10% of carriers. HTLV-2 has not been conclusively linked to any disease, but is sporadically associated with myelopathy and other neurologic disorders (1, 2). While HTLV-1 is endemic in Japan, the Caribbean, South America, and Central-Africa (3), HTLV-2 is more frequent among some Amerindian populations, some pygmy tribes in Africa, and persons who inject drugs (PWID) in North America and Europe (4).

The effects of HTLV co-infection on human immunodeficiency virus type 1 (HIV-1) have been widely studied. HTLV-1 co-infection has been associated with more rapid progression of HIV-1 disease, higher mortality, and increased frequency of opportunistic infections (2) but has also been associated with delaying HIV-1 disease progression (5). In contrast, HTLV-2 has been suggested to exert a negative effect on HIV-1 replication and has been associated with decreased mortality and slower HIV-1 disease progression (2, 5). Mechanisms underlying these various outcomes might partly be dependent on HTLV tropism for immune cells – HTLV-1 for CD4+ and HTLV-2 for CD8+ – as these types of T cells play key roles in HIV-1 infection.

Many of the Eastern European countries, including Estonia, have experienced concentrated HIV epidemics with the majority of HIV infections occurring among PWID (6). Estonia's capital Tallinn, a city of 440 000 residents with median age of 38 years, is estimated to have a PWID population of around 10 000, which accounts for approximately 70% of PWID in the country (7). As PWID are at risk for acquiring blood-borne infections, prevalences of some infections are well described. For example, co-infections with hepatitis viruses (HBV, HCV) have been shown to be high among PWID in Eastern European countries (8–11). In addition, previous reports have shown high prevalence of HTLV-2 among PWID (10%) (12–14) and HIV positive (HIV+) patients (15%) (15) in Western European countries. However, there are no reports describing the prevalence of HTLV-1/HTLV-2 among PWID in Estonia or Eastern Europe, where the HIV epidemic is continuing. Therefore, we aimed first to determine the prevalence and genotypes of HTLV-1/HTLV-2 among healthy volunteers and PWID in Estonia, and second to evaluate their associations with sociodemographic factors (age, gender), co-infections, and duration of persons who inject drugs (PWID).

Materials and Methods

The study included 345 PWID and 138 healthy volunteers. The PWID group was recruited by respondent-driven sampling method from a syringe exchange program in Tallinn,

Estonia, in 2011. The healthy volunteers group was collected to resemble the age distribution of Estonian general population. Sociodemographic data (age, gender) from healthy volunteers and PWID, and risk behaviour (duration of injection drug use (IDU), history of receptive needle sharing) from PWID were collected by self-reporting. Informed written consent was obtained from all the study subjects and the study was approved by the Ethics Committee of Tartu.

HIV, HCV, and HBV serostatus was determined as described previously (16). For the detection of HTLV-1/HTLV-2, DNA from 50 µl of peripheral blood mononuclear cells was extracted using Invitrogen PureLink Pro 96 Genomic DNA kit (Life Technologies, California, USA) according to the manufacturer's instructions. The long terminal repeat region of HTLV-1/HTLV-2 was amplified using nested PCR (17, 18), detailed conditions shown in Supplementary 1.

DNA extracted from chronically infected cell lines, MT-4 (HTLV-1+) and Gu (HTLV-2+) (19), served as positive controls in all PCR runs. PCR products were directly sequenced using the ABI Prism Big Dye 3.1 fluorescent terminator sequencing chemistry (Applied Biosystems, Foster City, CA) with the second round PCR primers.

Statistical analysis was performed by R 2.15.2 (www.r-project.org). Descriptive statistics (median and interquartile range/mean and standard deviations for continuous variables; percentages and absolute frequencies for categorical variables) were calculated. Differences between groups were explored by Fisher exact test or Mann-Whitney-Wilcoxon test, as appropriate. All statistical tests used a significance level of 0.05.

Results and Discussion

Of healthy volunteers 59 were men (43%) and median age was 37.5 years (interquartile range [IQR] 27–51 years). All healthy volunteers were negative for HIV, HCV, and HBV. Twenty eight percent (39/138) of healthy volunteers and 12% (43/345) of PWID were vaccinated against HBV. The studied PWID (n=345) were mainly male (79%) with median age of 30 years (IQR 25–34), and had prolonged duration of IDU (11 years; IQR 7–14). The rate of exposure to various blood-borne viruses was high: 50% were HIV+, 88% HCV+, 67% HBV+ (Table 1). Of PWID, 64% reported receptive needle sharing in lifetime and 18% at least once a month during last six months. These findings are consistent with previous studies of Estonian and other Eastern European PWID populations: primarily male subjects with extensive duration of IDU and high prevalence of exposure to blood-borne viruses (8–11).

All healthy volunteers (n=138) were negative for HTLV-1 and HTLV-2 (95% confidence interval [CI] 0.0–2.6). HTLV-1 was also not detected among PWID. We found one of the PWID to be positive for HTLV-2 (prevalence 0.3%; 95% CI 0.01–1.6), but we were unable to sequence it and therefore determine the HTLV-2 subtype (all HTLV positive control samples were sequenced successfully). The HTLV-2 positive subject was a 32-year old male with nine years of IDU, no reported history of receptive needle sharing, and HIV–, HCV+.

It is estimated that approximately 5–10 million individuals are carrying HTLV-1 infection worldwide (3). With HTLV-2 being less studied, estimations on the number of overall carriers is harder to make, but it has been estimated to be several million (4). In Central and Western Europe, the prevalence of HTLV-1, as well as HTLV-2, has been shown to be less than 0.1% among pregnant women and 0.005% among blood donors (20, 21). The prevalence of HTLV-1 is slightly higher in countries with high immigration from HTLV-1 endemic regions (mainly Portugal, Spain, France, UK) (22). In addition, higher frequencies of HTLV-2 have been described among PWID (around 3%) in Sweden (23), (around 10%) in Spain and Italy (12–14), and among HIV+ PWID (14.6%) in Ireland (15). In Eastern Europe, the data regarding the prevalences of HTLV-1 and HTLV-2 is largely absent except a single study from Romania where the prevalence of HTLV-1 was shown to be 0.053% among blood donors (20).

Similarly, the current study found HTLV-1 and HTLV-2 to be absent among healthy volunteers and nearly absent among PWID. While the situation among healthy volunteers in the current study is similar to that described in Western European countries, the finding of only a single case of HTLV-2 among PWID is somewhat surprising since HTLV-2 has been shown to be more prevalent among PWID in other European countries (12–15). This might be explained by three reasons. First, there are very few immigrants from HTLV-1/HTLV-2 endemic regions living in Estonia. Second, the PWID population in Estonia is relatively closed – even new HIV-1 strains have not spread and majority of infections are caused by CRF06_cpx since the outbreak in 2000 (24, 25). Third, the studied PWID were relatively young – whereas HTLV causes lifelong infection and the prevalence is usually higher among older people (3).

Our study has some limitations. First, the number of healthy volunteers included in the study was small. Hence, the result should be used cautiously in assessing the prevalence of HTLV in Estonian general population. Second, the prevalence of HTLV-1/HTLV-2 was determined by PCR analysis alone. As great majority of HTLV seropositive persons are also PCR positive (26), the true prevalence of HTLV in Estonia is in accordance with our results or only slightly underestimated.

In summary, we showed that HTLV is rare in Estonia. Since HTLV-1/HTLV-2 infections are rare among PWID in this region, it is unlikely that these viruses are influencing the course of HIV epidemics and disease progression in Estonia.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Highlights

- We examined 345 PWID and 138 healthy volunteers for HTLV-1 and HTLV-2 infection.
- We found all of the PWID and healthy volunteers to be negative for HTLV-1.
- We found one PWID to be HTLV-2 positive; all healthy volunteers were negative.
- All the figures were obtained by nested-PCR alone.

Table 1

Sociodemographic Factors and Co-Infection Statuses among Persons who inject drugs (PWID) and Healthy Volunteers in Estonia

	PWID (n = 345)	Healthy volunteers (n = 138)
Gender, n (%)		
Male	272 (79%)	59 (43%)
Age, median (IQR)		
	30 (25–34)	38 (27–51)
Duration of injection drug use, median (IQR)		
	11 (7–14)	NA
Receptively shared needles, n (%)		
Ever ^a	220 (64%)	NA
Frequently ^b	61 (18%)	NA
HIV status, n (%)		
HIV+	172 (50%)	0
HCV status, n (%)		
HCV+	306 (89%)	0
HBV status, n (%)		
HBV+	232 (67%)	0
HBV vaccinated	43 (12%)	39 (28%)
HTLV-1 status, n (%)		
HTLV-1+	0	0
HTLV-2 status, n (%)		
HTLV-2+	1 (0.003%)	0

Note.

^a reported receptive needle sharing in the past.

^b reported receptive needle sharing at least once a month during last six months.