

## Seroprevalence of HIV and HTLV in a representative sample of the Spanish population

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### SUMMARY

HIV and HTLV seroprevalence was determined by means of unlinked anonymous testing of 2144 sera, originally obtained from primary care patients by representative sampling of the Spanish population aged 15–39 years in 1996. HIV-1 seroprevalence was 4·3 per 1000 population in the 15–39 years age group [95% confidence interval (CI), 1·5–10·7] and 5·6 per 1000 (95% CI, 1·8–15·3) in the 20–39 years age group. Seroprevalence proved higher in males and urban residents. No antibodies to HIV-2 and HTLV-I were detected in any of the sera studied. However, presence of antibodies to HTLV-II was confirmed in one serum sample, while HTLV seroreactivity, though detected in another, could not be typed. The two HTLV-positive results equated to a seroprevalence of 1·9 per 1000 in the 20–39 years age group (95% CI, 0·3–8·6). HIV-1 seroprevalence was consistent with previous estimates yielded by back-calculation. The level of HTLV seroprevalence found suggests endemicity.

### INTRODUCTION

HIV infection is heterogeneously distributed throughout populations in line with risk practices, so that epidemiological surveillance is fundamentally conducted via sentinel populations [1]. Nonetheless, precise knowledge of the overall level of HIV infection in the population at large is of undeniable interest where public health and health care policy are concerned. While seroprevalence data obtained from different sentinel populations can prove extremely useful for specific preventive actions, they form a mosaic that is difficult to synthesize as a global seroprevalence figure [2]. In Spain, where health-care coverage is universal, primary care patients constitute

one of the sentinel populations most closely approximating the general population. Nevertheless, genuine population representativeness calls for suitable sampling.

The number of live infected persons in the global population can be estimated on the basis of HIV infection reporting systems [3], yet the necessary data are only available in some countries. The most widely used method for estimating the overall extent of HIV infection in the entire population is back-calculation based on reported AIDS cases [4]. Application of this method to Spain yielded an indirect estimate of 120 000 infected persons in 1991 (4·95 infected persons per 1000 population in the 15–59 years age group) [5]. However, in recent years the applicability of similar studies has become uncertain, owing to major changes

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witnessed in the natural course of HIV infection [4] and the revision of the AIDS case definition [6].

The present study sought to ascertain HIV-1 seroprevalence in a representative sample of the 1996 Spanish population aged 15–39 years. Furthermore, HIV-2, HTLV-I and HTLV-II antibody seroprevalence were also analysed.

## METHODS

A National Vaccine-Preventable Disease Survey was carried out in February 1996 [7]. To this end, a representative sample was obtained of the Spanish population, excluding the region of Catalonia, under the age of 40 years. Individuals were recruited from 123 blood-extraction centres in the public primary health-care network. Cluster sampling was conducted in three stages: (1) establishment of the number of sampling units (blood-extraction centres) in each region, in a manner proportional to the local population and urban-rural distribution; (2) random selection of participant centres in the respective regions; and (3) systematic random start sampling of patients in each such centre. Given the survey's stated objective, all immunodeficient patients, including persons with AIDS and HIV infection in advanced stages, were excluded. Epidemiological data were collected for each patient, and a blood specimen taken for determination of antibodies to the various vaccine-preventable diseases. When the study had been completed, all remaining sera were stored at  $-20^{\circ}\text{C}$ .

In 1998, on conclusion and independently of the above survey, it was proposed that the serum bank be used for the purpose of conducting an unlinked anonymous study into the seroprevalence of HIV-1, HIV-2, HTLV-I and HTLV-II antibodies. After due evaluation of the relevant technical and ethical aspects, the committee responsible for the serum bank approved the project. All sera corresponding to individuals aged 15–39 years were included. Sera aliquots were taken, along with the following information: sex, age group, urban (i.e. town of over 50000 inhabitants) or rural residence, and the respective weighting coefficient.

All sera were analysed at the national reference laboratory (National Microbiology Centre). For HIV diagnosis, the ELISA technique (Genelavia Mixt, Sanofi Diagnostics Pasteur SA, France) was used for screening, and a immunoblot assay (Chiron RIBA HIV-1/HIV-2 SIA, Chiron Corporation, USA) for confirmation of reactive sera and differentiation

between HIV-1 and HIV-2. Similarly, the HTLV-I and HTLV-II diagnostic strategy involved an ELISA (Murex HTLV I+II, Murex, UK) for screening and a immunoblot assay (Chiron RIBA HTLV-I/HTLV-II SIA, Chiron Corporation, USA) for confirmation and characterization.

In the calculation of seroprevalence, individuals were allocated weights that were inverse to their probability of selection, as a means of reconstructing the proportionality of the population. Taking the effect of cluster sampling into account, a Poisson distribution was used to obtain 95% confidence intervals (CI). Association between variables was measured by means of weighted prevalence odds ratios (OR). Analyses were performed with the aid of the Sudaan 7.0 and Epi-Info 6 computer software packages.

## RESULTS

The serum bank contained 2166 sera from persons aged 15–39 years, and of these, 22 could not be analysed for want of a sufficient quantity. Of the 2144 sera analysed, 6 were confirmed as HIV-1 positive. Once the proportionality of the sample had been reconstructed, seroprevalence rose to 4.3 per 1000 population in the 15–39 years age group (95% CI, 1.5–10.7).

Bearing in mind that the 15–39 years age range is very heterogeneous *vis-à-vis* HIV-1 infection (since it includes adolescents and adults) and that no infection was detected in the 15–19 years age group, subsequent analyses centred on the 20–39 years age group. Table 1 shows the results for 1637 sera of persons aged 20–39 years, 6 of whom were HIV-1 positive (5.6 per thousand; 95% CI, 1.8–15.3). Seroprevalence was higher in males than in females (OR = 3.1), and in the 20–29 than in the 30–39 years age group. Urban residents had a seroprevalence of 10.0 per 1000 *vs.* 1.4 per 1000 for rural residents (OR = 7.3). Seroprevalence in male urban residents aged 20–39 years rose to 17.4 per 1000. In no case however did these differences reach statistical significance.

None of the 2144 sera tested HIV-2 positive. Although neither the design nor the size of the sample was optimal for the study of HIV-2 seroprevalence in the population, these results reflect a very low prevalence (point estimate of zero per 1000 population aged 15–39 years; 95% CI, 0–1.7).

Presence of HTLV-II antibodies was confirmed in 1 of the 2144 sera analysed. Another serum, which had

Table 1. *HIV-1 seroprevalence among the population aged 20–39 years (Spain, 1996)*

	Number of sera analysed	HIV-1 positive sera	HIV-1 seroprevalence (per 1000 inhabitants)*	95% confidence interval*
Sex				
Males	469	4	8.3	2.5–23.9
Females	1168	2	2.7	0.3–10.2
Age (years)				
20–29	1077	4	6.7	2.3–18.0
30–39	558	2	4.3	0.6–17.8
Setting				
Urban	835	5	10.0	2.9–29.8
Rural	802	1	1.4	0.04–8.6
Total	1637	6	5.6	1.8–15.2

\* Seroprevalence and confidence interval estimates taking into account the effect of sampling.

tested HIV-1 positive, met the criteria for HTLV seropositivity but could not be typed, failing to react to HTLV-I gp46- and HTLV-II gp52-derived synthetic peptides. The single HTLV-II positive result yielded a prevalence of 0.6 per 1000 population in the 20–39 years age group (95% CI, 0.04–4.1), which rose to 1.9 per 1000 when both HTLV positive results were considered (95% CI, 0.3–8.6).

## DISCUSSION

The results suggest a 1996 HIV-1 seroprevalence of 5.6 per 1000 population in the 20–39 years age group, with odds of HIV infection being three times higher in males *vs.* females and seven times higher in urban *vs.* rural settings. Although incorporation of the cluster sampling effect results in wide confidence intervals, rendering it impossible to ascertain significant differences in the above variables, point estimates of seroprevalence and the epidemiological pattern described are nevertheless in line with existing epidemiological information on HIV and AIDS in Spain [5, 8].

The sample used was, in many ways, ideal for estimating HIV-1 seroprevalence in the Spanish population. The sampling was designed to ensure population representativeness and was based on the public primary care network which covers the entire population. The sample was obtained in a way that was divorced from the HIV infection status and risk practices of the individuals concerned, save for the exclusion of patients with manifest immunodeficiency. Notwithstanding this, generalization of the results to the Spanish population as a whole might be affected

by certain limitations. Firstly, the survey refers solely to young adults and adolescents. Nonetheless, it is precisely in these ages that most HIV-1 infections are concentrated; hence, 80.7% of AIDS cases in Spain are aged 20–39 years at date of diagnosis [9]. Secondly, the results, though applicable to Spain, exclude Catalonia, since this region did not take part in the Vaccine-Preventable Disease Seroprevalence Survey [7]. In the overall AIDS case count for Spain, Catalonia represents 22.1% of the total [9]. Thirdly, the prison population enjoys a specific health-care network and is, thus, not represented in the sample. Spanish prisons have an HIV diagnostic and control programme in place, with approximately 8000 infected inmates officially listed in 1996 [10]. Fourthly, immunodeficient persons were excluded from the initial sample, meaning that AIDS patients or individuals with advanced-stage HIV infection are therefore not represented. Based on AIDS registry data, Spain was estimated to have 15000 live AIDS cases at the end of 1996 [11]. Fifth and lastly, there is a possible bias inherent in the reduced likelihood of recruiting the healthiest subjects from health-care centres, the effect of which on HIV-1 seroprevalence estimation is difficult to quantify.

The sample size was considered acceptable for the study of HIV-1, but proved wanting for the study of the other human retroviruses. Earlier studies had found few cases diagnosed with HIV-2 infection in Spain, the majority being in persons from countries where this was endemic [12]. The absence of HIV-2 positive sera in the present survey is evidence of limited circulation of this virus. The two HTLV

positive sera denote a certain endemicity of this infection in Spain, probably due to its spread among injecting drug users [13, 14].

On the basis of the seroprevalence estimated in the present survey, an approximation can be made of the number of live persons who were infected with HIV-1 in Spain in 1996. A seroprevalence of 5.6 per 1000 applied to the Spanish population aged 20–39 years (excluding Catalonia) would give an estimate of some 60000 HIV-1 infected persons in this age range. In order to compute the total of HIV-1 infections in Spain, one would then have to apply the adjustments corresponding to cases under 15 and over 39 years old, residents of Catalonia, prison inmates, and patients with AIDS or in advanced stages of infection. On correction, the resulting estimate gives a range of 110000–140000 live persons infected with HIV-1.

The results of this study provide the first direct population-based estimates of overall HIV-1 seroprevalence in Spain, which support estimates obtained by indirect methods, and constitute a reference for future studies.

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