

# Seroepidemiological Survey of Human T-Lymphotropic Retrovirus among Indigenous Populations in Taiwan

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The prevalence of human T-lymphotropic retrovirus (HTLV) was examined in Taiwan's indigenous populations. In all, 797 healthy subjects in Taiwan including Han Chinese and nine indigenous populations (Ami, Atayal, Bunun, Saisiat, Paiwan, Puyuma, Rukai, Tsuo, and Yami) were examined for the presence of antibodies to HTLV by particle agglutination, indirect immunofluorescence and Western blot test. Two seropositive cases were found in this screening. One Saisiat male and a Han Chinese female were seropositive for HTLV. The Western blot profile indicated the virus was type-1 HTLV.

Human T-lymphotropic retrovirus (HTLV)<sup>1</sup> has been considered as the causative agent of adult T-cell leukaemia (ATL)<sup>2</sup> and HTLV-1 associated myelopathy (HAM/TSP).<sup>3</sup> HTLV showing endemic and non-endemic patterns of infection among human populations is not a common virus in man.

Taiwan is situated between HTLV-endemic Ryukyu (Southwestern part of Japan)<sup>4,5</sup> and non-endemic mainland China.<sup>6</sup> Recent seroepidemiological studies showed several seropositive cases of HTLV infection in the Taiwanese population.<sup>7,8</sup> In addition patients with ATL and HAM/TSP have been identified.<sup>8,9</sup> The Taiwanese population consists of Han Chinese and indigenous populations. The Han Chinese migrated from Fujian and Canton in mainland China several hundred years ago and are to be found throughout the island. The indigenous Taiwanese who live in the mountainous areas, on the southeastern coastal region, and on an island between Taiwan and the Philippines (Figure 1), have been classified into

nine ethnoculturally distinguishable populations. Ethnological data suggest that there is a close relationship between the indigenous Taiwanese and southeast Asian native groups.<sup>10</sup> Data from two nationwide seroepidemiological studies of HTLV are available<sup>7,8</sup> and both report positive cases in the Han Chinese with prevalence rates ranging from 0% to 1%. However, there is controversy over the presence of virus carriers in the indigenous populations. In one study seropositivity was reported in the north<sup>6</sup> whereas in another no seropositive cases were found among eight indigenous population groups.<sup>8</sup>

Because previous surveys did not cover all indigenous population groups, a serological survey of HTLV infection among all nine indigenous populations of Taiwan (Ami, Atayal, Bunun, Paiwan, Puyuma, Rukai, Saisiat, Tsuo, and Yami) was carried out.

## MATERIALS AND METHODS

### Population

According to the 1990 census, the indigenous population of the nine ethnic groups is about 340 000. The population size of each ethnic group is 130 000 in Ami, 80 000 in Atayal, 60 000 in Paiwan, 40 000 in Bunun, 8000 in Puyuma, 8000 in Rukai, 5000 in Tsuo, 4000 in

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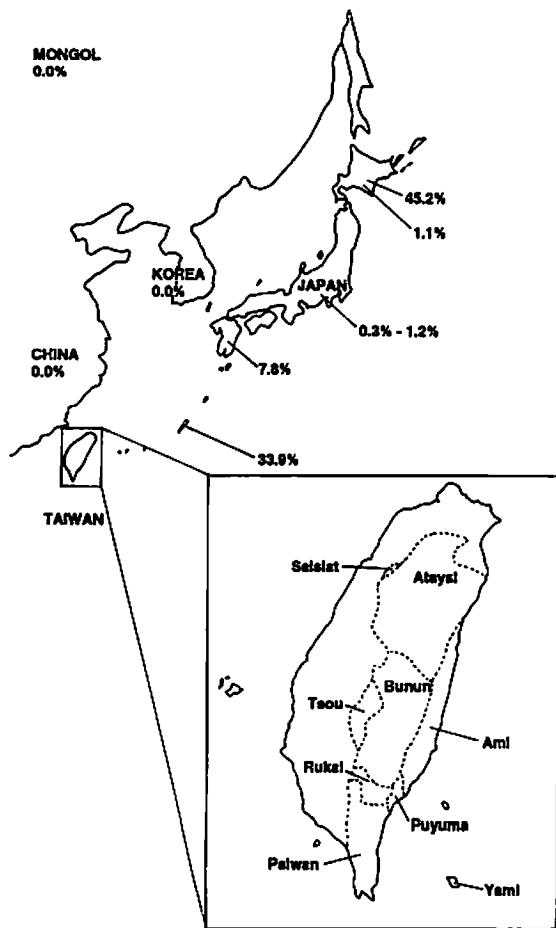


FIGURE 1 Location of indigenous populations in Taiwan and geographical distribution of HTLV-1 in the neighbouring countries of Taiwan based on data obtained in the previous<sup>6,15-17</sup> and unpublished (Mongol) studies. Numbers indicate percentages of seropositive adults. In Japan, the prevalence differs by the population and the Ainu (45.2%) and the Ryukyuan (33.9%) constitute the high-prevalence groups

Yami, and 4000 in Saisiat. They are principally agricultural workers and the Yamis also depend on fishing.

#### Sera Sampling

The populations were sampled for virological and genetic studies. In all, 797 blood samples were collected between 1989 and 1991 from healthy people of nine different indigenous populations and additionally from Han Chinese in Taiwan. The Ami, Bunun, and Paiwan which have comparatively large populations were sampled from two different villages. In Atayal, we sampled the southern population since the northern population had been studied previously.<sup>7</sup> The samples

were collected from volunteers and, using family records, those who were of mixed race were excluded. To be representative of the population and to avoid duplicate samples from the same family, only one member of any family was sampled. The donor's sex, age group, and indigenous population group are listed in Table 1 and their locations are mapped in Figure 1. All blood samples were collected from healthy individuals and were separated and stored at  $-80^{\circ}\text{C}$  before use. The origin and family history of the donors were recorded.

#### Detection of Antibodies

Antibodies to HTLV were screened by a particle agglutination (PA) test (Serodia HTLV, Fujirebio, Japan) which was improved to reduce false positives.<sup>11</sup> Reactions were examined at 1:16 dilution. Samples showing positive reactions at first screening were re-tested by PA test at further dilutions. In addition immunofluorescence (IF)<sup>12</sup> and Western blot (WB) (Problot HTLV, Fujirebio, Japan) testing was carried out. Also all samples were tested for antibodies to HIV-1 by PA test (Serodia HIV, Fujirebio, Japan).

#### RESULTS

Among Taiwan indigenous inhabitants, one male from Saisiat aged 56 years was seropositive in every test. The antibody titre in PA and IF tests was 512 and 20, respectively. Positive bands in the WB test were observed against p19, p24, p53 and gp46. He was born in an isolated Saisiat village in the mountainous area in the middle of Taiwan and had no risk factors for HTLV infection, such as drug abuse and blood transfusion. His wife, aged 55 years, born in the same village but from a different family, was negative. Of the 132 Han Chinese, one female aged 45 years was seropositive by PA, IF and WB tests. Antibody titres by PA and IF were 4096 and 20, respectively. Strong positive bands in the WB test were observed against p19, p24, p53, and gp46. Her forebears came from Fujian in mainland China and she also had no risk factors for HTLV infection. No difference in the pattern of antibody reactivity to viral antigens was observed between two positive cases. The intensity of positive reaction against p19 was stronger than to p24 in both cases.

All the plasma samples tested were negative for HIV-1 antibody.

#### DISCUSSION

Two out of 800 individuals were found to be HTLV seropositive. This low prevalence confirmed previous reports<sup>7,8</sup> and suggested that HTLV is not a major public health hazard in Taiwan. However, patients

TABLE 1 Age, sex and indigenous population group of sample

Population Age group (years)	Paiwan		Rukai		Atayal		Saisiat		Tsuo	
	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female
0-19	0	0	12	20	1	3	0	2	0	0
20-39	3	15	5	5	11	24	2	5	3	11
40-59	8	14	6	5	14	27	19	14	11	23
≥60	6	14	1	1	7	14	11	11	13	22
Total	17	43	24	31	33	68	32	32	27	56

Population Age group (years)	Yami		Puyuma		Ami		Bunun		Han Chinese	
	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female
0-19	31	47	1	0	0	0	1	1	0	6
20-39	0	0	0	7	6	5	11	19	13	82
40-59	0	0	9	20	8	17	9	19	8	23
≥60	0	0	15	12	19	17	13	15	0	0
Total	31	47	25	39	33	39	34	54	21	111

with ATL or HAM have been identified<sup>8,9</sup> and we should continue to monitor the epidemiology of HTLV.

According to Wiktor *et al.*<sup>13</sup> HTLV-1 and HTLV-2 can be distinguished by antibody reactivity in WB test. They state that the intensity of serum reactivity to p19 is stronger than to p24 in HTLV-1 infection. The two seropositive cases in this study suggested the type of HTLV in Taiwan to be type-1.

One of the nine indigenous populations, Saisiat, had not previously been examined for HTLV infection. We found one seropositive carrier in this group (1/64). A previous study<sup>7</sup> reported a seropositive married couple of Taiwan aborigines (an indigenous group in this study), however, the name of the tribe was not stated. This couple was thought to be from the Atayal because of the area of origin. No HTLV carrier has been found in the other seven indigenous populations so far.

The other HTLV carrier in this study was Han Chinese (1/132). Han Chinese are known to have a low prevalence of HTLV carriers (0-1%).<sup>7,8</sup> Taking our results with those of other serological studies, the origin of HTLV-1 in Taiwan is still not clear. Considering the relation between the Taiwanese population and its surrounding populations, several HTLV-1 introduction routes, such as via mainland China, Ryukyu, and or the Philippines,<sup>14</sup> are possible. Further

seroepidemiological surveys in China and in the Philippines are required and the comparative study of the HTLV-1 proviral genomes of Ryukyu, Taiwan, Han Chinese and indigenous inhabitants may help to answer this question. Initially genetic studies of the Taiwanese population were started and these are being expanded to isolate HTLV-1 proviruses from seropositive carriers.

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#### REFERENCES

- Poiesz B J, Ruscetti F W, Gazdar A F, Bunn P A, Minna J D, Gallo R C. Detection and isolation of type C retrovirus particles from fresh and cultured lymphocytes of a patient with cutaneous T-cell lymphoma. *Proc Natl Acad Sci USA* 1980; 77: 7415-19.
- Takatsuki K, Uchiyama T, Sagawa K, Yodoi J. Adult T-cell leukemia in Japan. In: Seno S, Takaku F, Irino S (eds). *Topics in Hematology* Amsterdam; Excerpta Medica, 1977, pp. 73-77.
- Osame M, Usuku K, Izumo S *et al.* HTLV-1 associated myelopathy, a new clinical entity. *Lancet* 1986; i: 1031-32.

- <sup>4</sup> Ishida T, Hinuma Y. Origin of Japanese HTLV-1. *Nature* 1986; **322**: 504.
- <sup>5</sup> Morofuji-Hirata M, Kajiyama W, Nakashima K *et al*. Prevalence of antibody to human T-cell lymphotropic virus type I in Okinawa, Japan, after an interval of 9 years. *Am J Epidemiol* 1993; **137**: 43-48.
- <sup>6</sup> Zeng Y, Lan X Y, Fang J *et al*. HTLV antibody in China. *Lancet* 1984; **i**: 799-800.
- <sup>7</sup> Kuo T T, Chan H L, Su I J *et al*. Serological survey of antibodies to the adult T-cell leukemia virus-associated antigen (HTLV-A) in Taiwan. *Int J Cancer* 1985; **36**: 345-48.
- <sup>8</sup> Pan I H, Lin C Y, Komoda H, Imai J, Hinuma Y. Seroepidemiology of adult T-cell leukemia virus infection and analysis of seropositive cases in Taiwan. *Proc Natl Sci Council B* 1986; **10**: 254-62.
- <sup>9</sup> Wang S J, Liu H C, Chi C W, Liu T Y, Chen P M. HTLV-1 associated myelopathy: first case report in Taiwan. *Acta Neurol Scand* 1991; **84**: 256-58.
- <sup>10</sup> Liu M C. *Culture and Art of the Formosan Aborigines*. Taipei: Hsiung-Shih Book, 1988, pp. 26-29.
- <sup>11</sup> Fujino R, Kawato K, Ikeda M, Miyakoshi H, Mizukoshi M, Imai J. Improvement of gelatin particle agglutination test for detection of anti-HTLV-1 antibody. *Jpn J Cancer Res* 1991; **82**: 367-70.
- <sup>12</sup> Hinuma Y, Nagata K, Hanaoka M *et al*. Adult T-cell leukemia: Antigen in an ATL cell line and detection of antibodies to the antigen in human sera. *Proc Natl Acad Sci USA* 1981; **78**: 6476-80.
- <sup>13</sup> Wiktor S Z, Alexander S S, Shaw G M *et al*. Distinguishing between HTLV-I and HTLV-II by western blot. *Lancet* 1990; **335**: 1533.
- <sup>14</sup> Ishida T, Yamamoto K, Omoto K. A seroepidemiological survey of HTLV-1 in the Philippines. *Int J Epidemiol* 1988; **17**: 625-28.
- <sup>15</sup> Hinuma Y, Chosa T, Komoda H *et al*. Sporadic retrovirus (ATLV) — seropositive individuals outside Japan. *Lancet* 1983; **i**: 824-25.
- <sup>16</sup> Maeda Y, Furukawa M, Takehara Y *et al*. Prevalence of adult T-cell leukemia virus-carriers among volunteer blood donors in Japan: a nation wide study. *Int J Cancer* 1984; **33**: 717-20.
- <sup>17</sup> Ishida T, Yamamoto K, Omoto K, Iwanaga M, Osato T, Hinuma Y. Prevalence of a human retrovirus in native Japanese: evidence for a possible ancient origin. *J Infect* 1985; **11**: 153-57.

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