

Survey of Anti-human T-Cell Leukemia Virus Type I Antibody in Family Members of Patients with Adult T-Cell Leukemia

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To evaluate the intrafamilial clustering of HTLV-I, we examined the sera or plasma of 296 healthy family members of patients with adult T-cell leukemia (ATL) for anti-HTLV-I antibodies. Of 296 subjects, 132 (44.6%) had anti-HTLV-I antibodies. Fifty-nine (41.0%) out of 144 males and 73 (48.0%) out of 152 females were seropositive. The positive rates of antibody to HTLV-I increased with age, especially between the 30-39 and the 40-49 age groups. Five out of 6 fathers, 3 out of 4 mothers, 31 (60.8%) out of 51 spouses, 40 (63.5%) out of 63 siblings and 46 (33.8%) out of 136 children of patients with ATL had anti-HTLV-I antibodies. Of 74 children with an ATL father, 14 (18.9%) were seropositive, while 32 (51.6%) out of 63 children with an ATL mother were seropositive. This difference was statistically significant ($P < 0.001$). Of those children with an ATL father, 12 (26.1%) out of 46 whose mothers were HTLV-I carriers had antibodies to HTLV-I. In contrast, none of the 13 children whose mothers were not carriers were seropositive. These results supported the hypothesis that the mother-to-child transmission is one of the most important modes of HTLV-I transmission. In wives of male patients with ATL, the positive rate of antibody to HTLV-I was 65.6% (21/32), and in husbands of female patients, it was 52.6% (10/19). The high positive rate of antibody to HTLV-I not only in wives of male patients but also in husbands of female patients suggests that either HTLV-I is more frequently transmitted from wives to their husbands than we had originally expected, or that ATL may develop even in wives who acquire HTLV-I from their husbands after marriage.

Key words: Adult T-cell leukemia — HTLV-I — Viral transmission — Family study

Human T-cell leukemia virus type I (HTLV-I)¹⁻³ is a causative agent of adult T-cell leukemia (ATL),^{4,5} a new clinical entity of peripheral T-cell malignancy, distributed in southwestern Japan. The same virus is related to a spastic paraparesis syndrome, called HTLV-I associated myelopathy (HAM).⁶ HTLV-I carriers exist in endemic areas of ATL,^{7,8} and about 0.05-0.1% of these carriers get ATL annually.⁹⁻¹¹ Natural transmission modes of HTLV-I are considered to be from mother to child and between spouses, especially from husband to wife.¹²⁻¹⁴ While there have been many reported cases of ATL with a familial disposition,^{11,15} to date it has not been reported that natural horizontal transmission of HTLV-I between spouses may induce an overt manifestation of ATL. In healthy family members of patients with ATL, the prevalence of HTLV-I carriers is very high.^{11,15,16} However, detailed data about HTLV-I transmission in families of patients with ATL are scarce. To evaluate the intrafamilial clustering of HTLV-I and to explore the characteristic HTLV-I transmission modes in families of patients with ATL, we examined the sera

or plasma of healthy family members for antibodies to HTLV-I.

MATERIALS AND METHODS

Two hundred and ninety-six healthy family members (male 144, female 152, 8-86 years old) of 77 patients (male 44, female 33) with ATL, who were seen from 1982 to 1989, were examined for antibodies to HTLV-I in their sera or plasma. The details are shown in Table I.

Sera or plasma samples obtained from family members were diluted 10-fold and antibodies to HTLV-I were determined using the indirect immunofluorescence method utilizing MT-1, MT-2 and SLB-1, which are HTLV-I-infected cell lines, as target cells. Chi-square tests were used to analyze the data.

RESULTS

Of 296 subjects, 132 (44.6%) had anti-HTLV-I antibodies in their sera or plasma. Fifty-nine (41.0%) out of 144 males and 73 (48.0%) of 152 females were seropositive. The comparison by relationship is shown in Table I.

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Age- and sex-specific positive rates of antibody to HTLV-I are shown in Fig. 1. Positive rates increased with age. There was an acceleration of rate between the 30-39 and the 40-49 age groups. In the over-60 age

group, the positive rate in females was higher than in males.

Parents (Table II) Five out of 6 fathers of patients with ATL had anti-HTLV-I antibodies. Three out of 4 mothers of patients with ATL had HTLV-I antibodies. It is interesting to note that there was one seronegative mother.

Table I. Positive Rates for Anti-HTLV-I Antibody in Family Members of ATL Patients

	No.	Anti-HTLV-I antibody-positive	%
Parents	10	8	80.8
Spouses	51	31	60.8
Siblings	63	40	63.5
Children	136	46	33.8
Others	36	7	19.4
Total	296	132	44.6

Table II. Anti-HTLV-I Antibody in Parents of ATL Patients

Sex of ATL patient	Father		Mother	
	Anti-HTLV-I antibody (+)	Anti-HTLV-I antibody (-)	Anti-HTLV-I antibody (+)	Anti-HTLV-I antibody (-)
Male	5	1	2	1
Female			1	
Total	5	1	3	1

Table III. Positive Rates for Anti-HTLV-I Antibody in Married Couples
(A) Spouses of ATL patients

Sex of ATL patient	Anti-HTLV-I antibody		Total
	(+)	(-)	
Male (wife)	21 (65.6%)	11	32
Female (husband)	10 (52.6%) ^{a)}	9	19
Total	31 (60.8%)	20	51

a) High prevalence of HTLV-I carriers in husbands of female patients with ATL is inexplicable in term of the increase of couples who are both anti-HTLV-I antibody-positive by transmission from husband to wife, because it is considered that ATL does not develop in HTLV-I carriers who have acquired the virus in adulthood.

(B) Subjects over 40 years of age in Tsushima. Comparison by spouse's positivity^{a)}

	Anti-HTLV-I antibody of spouse		Total
	(+)	(-)	
Female (wife)	63/99 ^{b)} (63.6%)	74/473 (15.6%)	137/572 (24.0%)
Male (husband)	70/146 (47.9%) ^{c)}	37/458 (8.1%)	177/604 (17.7%)

a) This table is taken from a report by Tajima *et al.*²⁶⁾

b) Anti-HTLV-I antibody-positive/tested.

c) High prevalence of HTLV-I carriers in husbands with an HTLV-I carrier wife is due to the increase of couples who are both anti-HTLV-I antibody-positive by transmission from husband to wife.

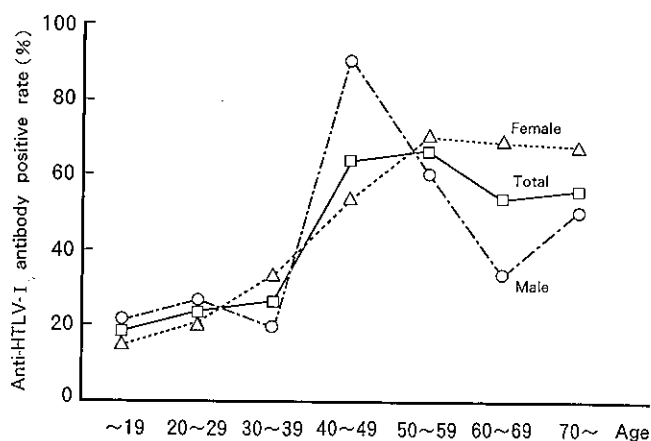


Fig. 1. Age- and sex-specific positive rates for anti-HTLV-I antibody in families of ATL patients. The positive rates increased with age, especially between the 30-39 and the 40-49 age groups. In the over-60 age group, the positive rate in females was higher than in males.

Table IV. Positive Rates for Anti-HTLV-I Antibody in Siblings of ATL Patients

Sex of ATL patients		Anti-HTLV-I antibody		Total
		(+)	(-)	
Male	Brothers	7 (41.2%)*	10	17
	Sisters	15 (62.5%)	9	24
	Total	22 (53.7%)	19	41
Female	Brothers	8 (100%)*	0	8
	Sisters	10 (71.4%)	4	14
	Total	18 (81.8%)	4	22
Total	Brothers	15 (60.0%)	10	25
	Sisters	25 (65.8%)	13	38
	Total	40 (63.5%)	23	63

* $P < 0.02$.

Table V. Positive Rates for Anti-HTLV-I Antibody in Children of ATL Patients

Sex of ATL patient		Anti-HTLV-I antibody		Total
		(+)	(-)	
Male	Sons	7 (16.7%)*	35	42
	Daughters	7 (21.9%)	25	32
	Total	14 (18.9%)**	60	74
Female	Sons	19 (57.6%)*	14	33
	Daughters	13 (44.8%)	16	29
	Total	32 (51.6%)**	30	62
Total	Sons	26 (34.7%)	49	75
	Daughters	20 (32.8%)	41	61
	Total	46 (33.8%)	90	136

* $P < 0.01$, ** $P < 0.001$.

Table VI. Positive Rates for Anti-HTLV-I Antibody in Children with an ATL Father. Comparison by Mother's Positivity

Anti-HTLV-I antibody of mothers		Anti-HTLV-I antibody		Total
		(+)	(-)	
(+) Mother	Sons	6 (22.2%)	21	27
	Daughters	6 (31.6%)	13	19
	Total	12 (26.1%)	34	46
(-) Mother	Sons	0 (0%)	7	7
	Daughters	0 (0%)	6	6
	Total	0 (0%)	13	13

Spouses (Table III) Twenty-one (65.8%) out of 32 wives of male patients with ATL had anti-HTLV-I antibodies. Likewise, a high positive rate of antibody to HTLV-I in husbands of female patients was found. Ten (52.6%) out of 19 husbands were seropositive.

Table VII. Positive Rates for Anti-HTLV-I Antibody in Subjects over 30 Years of Age in Tsushima^{a)}

	Tested	Anti-HTLV-I antibody-positive
Male	948	170 (17.9%)
Female	1634	415 (25.4%)
Total	2582	585 (22.7%)

a) The Tsushima Islands are one of the HTLV-I-endemic areas in Nagasaki Prefecture, Japan. This table is taken from a report by Tajima *et al.*²⁶⁾

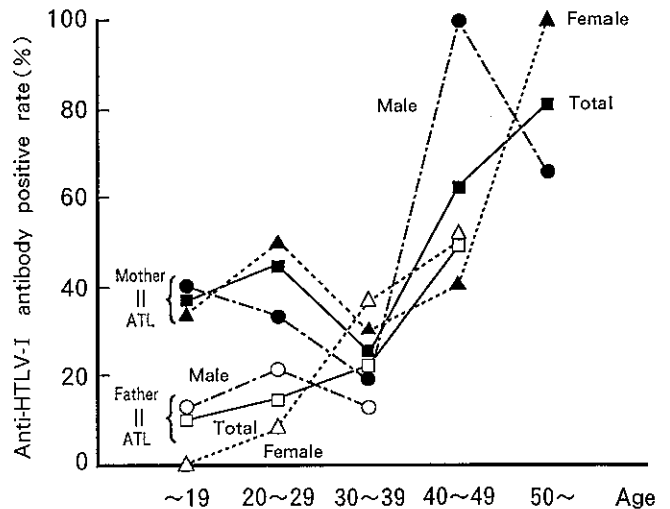


Fig. 2. Age- and sex-specific positive rates for anti-HTLV-I antibody in children of ATL patients. Children with an ATL mother had higher positive rates than those with an ATL father, in the under-30 age group. ○, male children with an ATL father; △, female children with an ATL father; □, all children with an ATL father; ●, male children with an ATL mother; ▲, female children with an ATL mother; ■, all children with an ATL mother.

Siblings (Table IV) The positive rate of antibody to HTLV-I in siblings of patients with ATL was 63.5% (40/63). In brothers of patients with ATL it was 60.0% (10/25) and in sisters it was 65.8% (25/38).

The positive rate of antibody to HTLV-I in siblings of female patients with ATL was slightly higher than in those of male patients. In siblings of female patients, 18 (81.8%) out of 22 subjects, and in those of male patients, 22 (53.7%) out of 41 subjects were seropositive. This difference was especially significant in brothers of patients with ATL. In brothers of female patients, all of the 8 subjects, and in brothers of male patients, 7 (41.2%) out of 17 subjects were seropositive ($P < 0.02$).

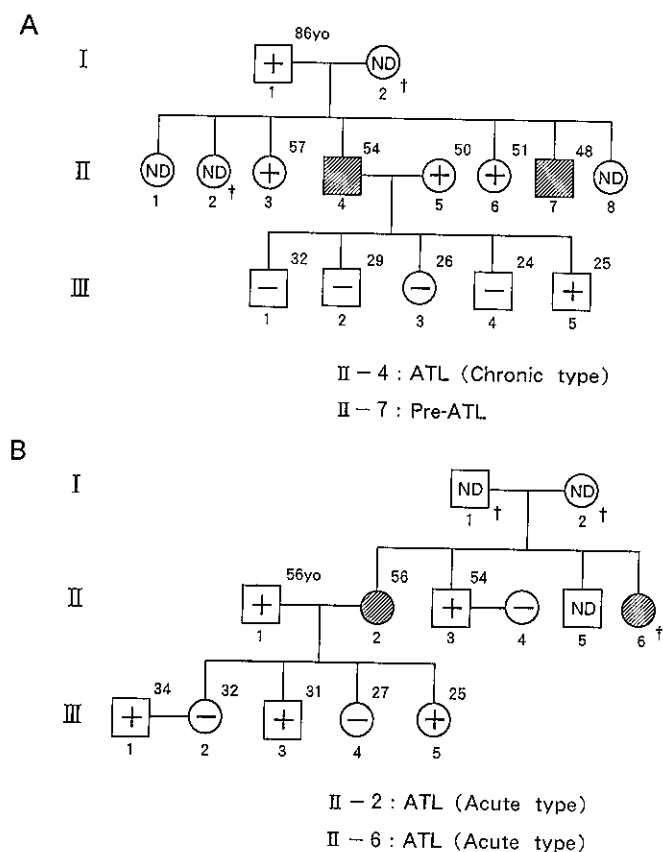


Fig. 3. Pedegrees of ATL patients and their relatives examined for anti-HTLV-I antibody. (A) A family that has one male with chronic-type ATL and his younger brother with pre-ATL. (B) A family that has two sisters with acute-type ATL. □, male; ○, female. ■, ●, ATL patient; ⊕, ⊕, anti-HTLV-I antibody-positive; ⊖, ⊖, anti-HTLV-I antibody-negative; ND, (ND), not done; †, †, died. Figures given as superscripts to the right of symbols indicate age.

Children (Tables V and VI) Forty-six (33.8%) out of 136 children of patients with ATL had anti-HTLV-I antibodies. The positive rate of antibody to HTLV-I was 34.7% (26/75) in sons, and 32.8% (20/61) in daughters. In children with an ATL father, 14 (18.9%) out of 74 subjects were seropositive. On the other hand, in children with an ATL mother, the positive rate of antibody to HTLV-I was significantly higher than in those with an ATL father, with 32 (51.6%) out of 62 subjects being seropositive ($P < 0.001$). In the former group, 12 (26.1%) out of 46 children with an HTLV-I carrier mother were seropositive. In contrast, none of the 13 children with a non-carrier mother was seropositive. Sex- and age-specific positive rates of anti-HTLV-I antibody are shown in Fig. 2. We found a tendency for children

under 30 years of age with an ATL mother to have higher positive rates than those with an ATL father.

Examples Fig. 3 shows examples of pedigrees of ATL families. One (A) is of a family that has one male with chronic-type ATL and his younger brother with pre-ATL.¹⁷⁾ The other pedigree (B) is of a family that has two sisters with acute-type ATL.

DISCUSSION

Because of the biological characteristics of HTLV-I, the presence of antibody to the retrovirus in serum means that HTLV-I infection has continued to exist. Therefore, an individual who has antibody to HTLV-I in his serum can be considered to be an HTLV-I carrier.¹⁸⁾ Several epidemiological surveys of anti-HTLV-I antibody in endemic areas have clarified that the modes of natural transmission of HTLV-I are vertically from mother to child, and horizontally between spouses, especially from husband to wife.¹²⁻¹⁴⁾ Since HTLV-I shows cell-associated infectivity and does not infect efficiently under cell-free conditions *in vitro*,¹⁹⁻²²⁾ transmission is likely to occur by transfer of provirus-containing cells. In other studies,^{23,24)} we have demonstrated that HTLV-I can be found in the T-cells in breast milk of HTLV-I carrier mothers. We further found that HTLV-I can be transmitted to the common marmoset by oral feeding of human milk gathered from HTLV-I carrier mothers. Thus, transmission of HTLV-I from mother to child can be explained to be via breast milk. Nakano *et al.*²⁵⁾ have demonstrated that HTLV-I infected T-cells existed in the semen of HTLV-I carrier males and suggested that transmission from husband to wife was via the semen. This study was designed to evaluate the intrafamilial clustering of HTLV-I and to explore the characteristic HTLV-I infection modes in families of patients with ATL.

As a whole, the positive rate of antibody to HTLV-I in family members of ATL patients was 44.6%, which was higher than the positive rate of anti-HTLV-I antibody found in the general population (22.7%)²⁶⁾ of the HTLV-I endemic area (Table VII). The positive rates of anti-HTLV-I antibody increased with age, especially between the 30-39 and the 40-49 age groups. The rates in females were higher than in males over 60 years of age. These trends have also been shown in the general population in endemic areas.^{7,8,10,13,14,26)}

The HTLV-I transmission rate from mother to child was estimated to be about 10-30%.^{12,26-28)} In this study, children under 19 years of age with an ATL mother had a slightly high positive rate (37.5%) of anti-HTLV-I antibody, in comparison with the transmission rate in HTLV-I carriers. This suggests that HTLV-I carrier mothers who may get ATL in the future are at high risk

of viral transmission. Further examination of this possibility is necessary.

In our study, the prevalence of HTLV-I carriers in children with an ATL mother was higher than in those with an ATL father. In the latter group, no children whose mothers were not HTLV-I carriers had antibody to HTLV-I. This indicates that vertical mother-to-child transmission is one of the most important modes of HTLV-I natural infection, not only in the general population but also in families of patients with ATL.

In siblings, especially in brothers of female patients with ATL, the prevalence of HTLV-I carriers was higher than in those of male patients. This finding is so far inexplicable. The most interesting finding in this study was that the prevalence of HTLV-I carriers was very high not only in wives of male patients with ATL but also in husbands of female patients. High prevalence of HTLV-I carriers in wives of male patients indicates that transmission of HTLV-I from husband to wife is very important as a natural infection mode. But, if it is true that no patient with ATL has developed from HTLV-I carriers to whom HTLV-I had been transmitted in adulthood, and that HTLV-I transmission in a married couple is almost entirely from husband to wife, the high prevalence of HTLV-I carriers in husband of female patients is difficult to explain in terms of these two main modes of HTLV-I infection. Our findings suggest the possibility of either reverse transmission of HTLV-I, i.e. from female

patients to their husbands, or the development of ATL in HTLV-I female carriers who acquired HTLV-I primarily from their husbands post marriage. Some reports^{16, 29-31)} indicated high positive rates of HTLV-I antibody in husbands of female patients with ATL, but the number of such cases was too small for reliable analysis. Further examination of a large number of husbands of female patients and their parents is necessary to investigate this further.

In conclusion, for both the general population in an endemic area, and families of patients with ATL, two transmission modes of HTLV-I i.e. from mother to child and from husband to wife seem to be very important. However, interesting exceptions were noted. In some family members of patients with ATL, it appears either that HTLV-I was transmitted from female patients to their husbands, or that ATL may develop even in wives who had acquired HTLV-I from their husbands. It is important that these two questions are investigated further.

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