ment in Table 2. The results of this experiment confirmed the results in Table 2, revealing a 70% decrease in the apparent rate of host protein synthesis prior to the onset of high levels of HTLV-IIIB protein synthesis and a return to "normal" rates of protein synthesis accompanying the synthesis of high levels of *gag* protein.
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Human T Cell Leukemia Viruses Use a Receptor Determined by Human Chromosome 17

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Human T cell leukemia viruses (HTLV-I and HTLV-II) can infect many cell types in vitro. HTLV-I and HTLV-II use the same cell surface receptor, as shown by interference with syncytium formation and with infection by vesicular stomatitis virus (VSV) pseudotypes bearing the HTLV envelope glycoproteins. Human-mouse somatic cell hybrids were used to determine which human chromosome was required to confer susceptibility to VSV(HTLV) infection. The only human chromosome common to all susceptible cell hybrids was chromosome 17, and the receptor gene was localized to 17cen–qter. Antibodies to surface antigens known to be determined by genes on 17q did not block the HTLV receptor.

HTLV-I IS ETIOLOGICALLY ASSOCIATED with adult T cell leukemia-lymphoma (*1*) and tropical spastic paraparesis (*2*), while HTLV-II is associated with T cell hairy leukemia (*1, 3*). Although only T cells appear to be sensitive in vitro to transformation (immortalization) by HTLV (*4*), many human and mammalian cells can be infected including sarcoma cell lines, and epithelial and endothelial cells (*5, 6*). The only retroviral receptor molecule unequivocally identified to date is the CD4 leukocyte antigen, which is used by human and simian immunodeficiency viruses (*7*).

For HTLV-I and HTLV-II, cell surface receptors can be detected by assays of syncytium induction (*6*), virion binding (*8*), and VSV pseudotypes (*9*). VSV(HTLV) pseudotypes acquire the host range and receptor specificity of HTLV detected by VSV plaque formation. Cells chronically producing retroviruses express viral envelope glycoproteins that mask or down-modulate receptor expression at the cell surface. We made use of this phenomenon, known as receptor interference (*1, 7, 9*) to confirm that HTLV-I and II used a common receptor and investigated whether its determining gene or genes could be mapped.

Human and hamster cells expressed HTLV-I and II receptors, whereas mouse cells, the bovine MDBK line, and the rat NRK line were relatively resistant (Table 1). Human cells productively infected with the PL and PK isolates of HTLV-I (HOS/PL, HT1080/PK) showed receptor interference with pseudotype plating and syncytium induction of HTLV-I and HTLV-II. From these results and previous findings with cat cells (*9*), we conclude that HTLV-I and HTLV-II bind to the same receptor, whereas bovine leukemia virus (BLV) uses a distinct receptor. This accords with the antigenic relation (*9, 10*) between the gp46 outer envelope glycoprotein of HTLV-I and II, which is thought to interact with the receptor.

Since plating efficiency of VSV(HTLV) on murine cells was approximately 1% that of human cells, we used human-mouse somatic cell hybrids to assign the receptor gene or genes to a human chromosome. VSV pseudotypes have previously been used for the assignment of retrovirus receptor genes to mouse chromosomes (*11*) and to human chromosome 19 (*12*). The only human chromosome common to each of the seven sensitive hybrid cell lines was chromosome 17 (Table 2). Among the fifteen insensitive hybrid lines, however, two contained human chromosome 17. The presence of chromosome 17 in these two hybrids was confirmed by expression of the MIC6 cell surface antigen detected by flow cytometry with monoclonal antibody H207 (*13*). The receptor gene in these hybrids may either be deleted or not expressed. Two cell hybrids, PCTBA1.8 and GPT17/3, were independently derived and carried chromosome 17 as the only human genetic contribution (*13, 14*). These hybrids were sensitive to both VSV(HTLV-I) and VSV(HTLV-II). The gene determining the HTLV receptor can therefore be assigned to human chromosome 17.

The sensitivity of human-mouse somatic hybrids containing fragments of chromosome 17, as shown in Fig. 1, was tested to obtain a regional localization of the receptor gene. Since the hybrid P7A/2 (*15*) lacking the whole of the short arm of chromosome 17 other than the centromeric region, was sensitive to VSV(HTLV-I and II), the receptor gene can be localized to 17cen–qter. Furthermore, selection against chromosome 17 in this hybrid (*15*) conferred resistance to these pseudotypes. Two hybrids, PJT2A1

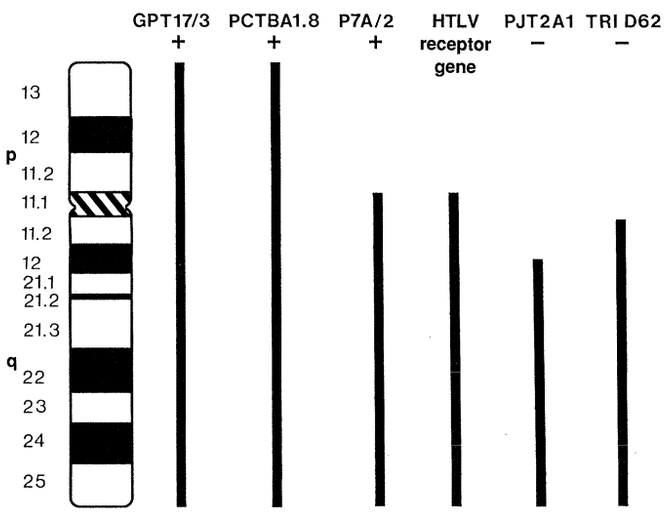

Fig. 1. Regional localization of the HTLV receptor gene on chromosome 17. VSV(HTLV-I) and VSV(HTLV-II) pseudotypes were plated on somatic cell hybrids (*14–16*) containing different portions of human chromosome 17. Susceptibility (+) and resistance (–) was scored as for Table 2.

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Table 1. Susceptibility of cells to VSV(HTLV) pseudotypes and to HTLV-induced syncytium formation. VSV was propagated in C91/PL T cells or HOS/PL osteosarcoma cells, which produce HTLV-I, and in C1812 T cells, which produce HTLV-II (MO strain), as described (*9*). The specific VSV(HTLV) pseudotype fraction was determined as the plaque-forming units (pfu) resistant to neutralization by hyperimmune anti-VSV immunoglobulin G (IgG) while remaining sensitive to human antiserum specific for HTLV-I or HTLV-II neutralization (*9*). Nonneutralized VSV and VSV(HTLV) pseudotypes were plated on monolayer cultures for 1 hour [after treatment with Polybrene (25 μg/ml) for pseudotypes], the monolayers were washed and overlaid with nutrient agar medium. Plaques were counted 40 to 48 hours later. Syncytial assays (*6*) were carried out by adding 5×10^5 C91/PL cells (HTLV-I) or C1812 cells (HTLV-II) to an equal number of HOS monolayer cells. After incubation overnight, the monolayers were washed, and the presence of syncytia was scored as positive (+) if >10% nuclei were in multinucleate giant cells. Control cultures and those with negative (−) scores had <2% nuclei in binucleate or multinucleate cells. ±, Heb7a cells were relatively resistant to virus-induced cell fusion. NT, not tested.

Cell line	Virus titer ($\log_{10}$ pfu/ml)			Syncytia		
	VSV	VSV(HTLV-I)	VSV(HTLV-II)	HTLV-I	HTLV-II	BLV
Human						
HOS	8.8	3.1	4.0	+	+	+
HOS/PL	8.9	<1.0	<1.0	−	−	+
HT1080	8.7	4.1	NT	+	+	+
HT1080/PK	8.8	1.0	1.0	−	−	+
RSb	NT	NT	NT	+	+	+
Heb7a	8.6	4.0	4.3	±	±	±
Hamster						
BHK21	9.2	3.3	NT	+	+	−
A23	8.2	9.1	NT	+	+	−
Murine						
LMTK$^-$	8.6	2.1	2.2	−	−	−
NIH 3T3 TK$^-$	9.0	2.1	1.3	−	−	−
PG19	8.5	<1.0	<1.0	−	−	−
PCC41RA	8.3	2.2	2.3	−	−	−
Rat						
NRK	9.0	<1.0	<1.0	−	−	−
XC	8.9	3.9	NT	+	+	−
Bovine						
MDBK	9.1	<1.0	NT	−	−	+

(*16*) and TRI D62 (*14*), were resistant to VSV(HTLV-I and II), suggesting a more precise localization of the receptor gene to the region 17cen–q12. However, because two hybrids retaining chromosome 17 (Table 2) were resistant to infection with VSV(HTLV-I), negative results should be treated with caution.

A number of cell surface molecules encoded by genes on human chromosome 17 suggest potential candidates for the HTLV receptor. These include *NGL* (also known as the c-*erb*B-2 or *neu* oncogene); *CD7*, a lymphocyte antigen; nerve growth factor receptor (*NGFR*), and *MIC6* (*17, 18*). Mouse cells transfected with and expressing the human *NGL* gene were resistant to infection with VSV(HTLV-I). Antibodies to *NGL*, *MIC6*, and *CD7* each failed to block HTLV-I–induced syncytial formation and pseudotype plating.

Transgenic mice expressing the *tat* gene of HTLV-I develop a syndrome resembling neurofibromatosis (*19*), and it is noteworthy that the gene determining von Recklinghausen neurofibromatosis has been located to human chromosome 17cen–q21 (*20, 18*). Although a link between HTLV *tat* and receptor gene expression is not immediately apparent, this may not be coincidental. The interleukin-2 receptor (IL-2R) has been suggested as an HTLV binding site (*21*); however, we found (*5*) that HTLV-I infects

Table 2. Susceptibility of somatic cell hybrids to infection by VSV(HTLV-I). Adherent mouse-human and rat-human (SIF) hybrid cells (*23*) were infected with VSV(HTLV-I) giving a titer of 10^4 pfu/ml on Heb7a cells. Hybrid cells were scored as positive (+) if the pseudotype titer was $>2 \times 10^3$ pfu/ml and as negative (−) if less than 2×10^2 pfu/ml. The chromosome content of the hybrid cells was ascertained by karyotype, isozyme, and antigenic analysis (*23*). The pseudotype sensitivity of control human Heb7a cells, the four lines of mouse cells, and the NRK rat cell line used for generating the somatic hybrids is shown in Table 1.

Cell line	Virus plating	Human chromosome																						
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	X	Y
MOG2E5	+	1		3	4	5	6	7	8	9	10		12	13	14	15	16	17	18			21	22	X	
MOG2C2	+	1		3	4	5		7	8	9	10	11			14	15	16	17	18	19	20	21	22	X	
PJTHP	+															15		17		19			22		
DUR4R4	+			3		5			8			11	12	13	14			17	18		20	21	22		
DUR4.3	+			3		5						11	12	13	14	15		17	18		20	21	22	X	
PCTBA1.8	+																	17							
GPT17/3	+																	17							
FIR5R3	−														14				18						
HORL411B6P2	−	1		3								11		13					18				22	X	
DT1.2.4.	−			3								11				15			18		20	21		X	
3E7	−																								Y
1W1LA4	−											11													
HORLI	−															15									
HORL9x	−																							X	
3W4C17	−							7			10	11	12		14	15		17				21		X	
MOG13/10	−	1		3													16					21	22	X	
THYB133	−																					21		X	
HORP9.5	−										10	11	12		14									X	
2860H7	−																16								
CTP34B4	−	1	2	3		5	6	7	8				12		14		16	17	18					X	
SIF15	−		2				6	7			10				14	15					20			X	
SIF4A31	−			3	4	5	6								14		16								
No. concordant		14	13	14	16	17	13	14	17	17	14	13	15	17	13	15	13	20	15	17	16	15	18	9	14
No. discordant		8	9	8	6	5	9	8	5	5	8	9	7	5	9	7	9	2	7	5	6	7	4	13	8

cells negative for Tac antigen (the α chain of IL-2R) encoded by chromosome 10 (*22*).

The chromosomal localization of the gene encoding the HTLV receptor gene will be useful for further elucidation of the receptor by immunological and genetic techniques.

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A DNA Binding Protein Regulated by IL-4 and by Differentiation in B Cells

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The class II (Ia) major histocompatibility complex antigens are a family of integral membrane proteins whose expression is limited to certain cell types, predominantly B lymphocytes, macrophages, and thymic epithelial cells. In B cells, Ia expression is both developmentally regulated and responsive to external stimuli. The differentiation of early B stem cells to mature B lymphocytes is accompanied by the appearance of cell surface Ia antigens; the transition to plasma cells results in loss of class II gene expression. In Ia-expressing B cells, the T cell–derived lymphokine interleukin-4 (IL-4) increases such expression by an as yet undefined mechanism. Chloramphenicol acetyltransferase gene expression was cis-activated by a region of the Ia $A\alpha^k$ gene in a B lymphoma line, but not in a myeloma line. A nuclear protein that bound to two sites within this region, upstream from previously described transcription elements, was found in normal spleen cells. This binding activity was also found in spleen extracts from athymic mice, which lack T lymphocytes, and in Ia-positive B lymphocyte tumor cell lines, demonstrating that it is a B cell protein. Further analysis showed the activity to be undetectable in an Ia-negative pre-B cell line and in three plasmacytoma cell lines that are Ia negative. IL-4 treatment of normal and athymic mouse spleen cells greatly increased the binding of this nuclear protein to these two sites, concomitant with increased MHC class II gene transcription. Thus, B cells contain a sequence-specific DNA-binding activity whose level is influenced both by IL-4 and by differentiation signals.

THE PRESENCE AND THE LEVEL OF cell surface class II major histocompatibility complex (MHC) gene products contribute to the intensity of an immune response [reviewed in (*1*)]. Tissue-specific and inducible control of transcription affect the level of class II MHC (Ia) gene expression (*2*). Because DNA regions upstream from several other class II MHC genes were active in cis (*3*), we studied a region upstream from the murine MHC Aα gene. A 1.3-kb Sau 3AI restriction fragment was cloned upstream from the SV40 promoter of pA10CAT (*4*). This construct (Fig. 1B) directed the synthesis of 70-fold more chloramphenicol acetyltransferase than the control vector, pA10CAT, when each was transfected (*5*) into the class II–positive B lymphoma M12.A2 (Fig. 1A). In the myeloma cell line XAg653, this DNA region provided no cis-activating effect (Fig. 1A). Transcriptional motifs, called X and Y, are conserved in all class II MHC genes (*6*) and are present at the downstream end of the Aα Sau 3AI fragment. Similar assays showed that the Aα X and Y sequences conferred little cis-activating effect in M12.A2 (*7*). Thus, the DNA region upstream from Aα (Fig. 1A) was active in cis in a B cell line but not in a myeloma cell line, and sequences other than the X-Y region appeared to contribute to cis-activation.

The activation of transcription by specific DNA sequences is often mediated by transcription factors that bind to these sequences. Therefore, we assayed nuclear extracts (*8*) by mobility shift analyses (*9*) with radiolabeled DNA fragments that represented the DNA within the 1.3-kb Sau 3AI fragment described above. For two restriction fragments (fragments I and II in Fig. 1C) in addition to the X-Y-containing fragment (IV in Fig. 1C), a retarded band was present when nuclear extracts from spleen cells were used (lanes 3 and 10 in Fig. 1D). Formation of these proteinase K–sensitive (*7*) complexes was blocked by competition with excess unlabeled probe DNA (lanes 4 and 11 in Fig. 1D), but not by several unrelated competitor DNAs (lanes 5, 6, 12, and 13). To determine whether B lymphocytes contain the binding activity, we prepared splenic nuclear extracts from athymic nude mice, whose spleens lack T cells and consist primarily of B cells. The factor was found in nude mice (lanes 7 and 14 in Fig. 1D). B lymphoma lines that express Ia constitutively were also assayed. Nuclear extract from M12.4.1, a class II–positive mature B cell lymphoma line, generated a DNA-protein complex that comigrated with the band from normal spleen cell extract (lanes 8 and 15).

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