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A human T-cell antigen receptor β chain gene maps to chromosome 7

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cDNA clones which encode the human and mouse T cell antigen receptor β chain gene have previously been isolated. We have used a mouse cDNA clone to map the chromosomal position of a human β chain gene. Southern blot analysis of DNA prepared from somatic cell hybrids has assigned this gene to chromosome 7. The use of a hybrid containing a chromosome 7 translocation has further localised this gene to the region 7q22-qter.

Key words: human T-cell antigen receptor/chromosomal localisation/chromosome 7

Introduction

The T-cell receptor for antigen has been identified in both mouse and man as a 90-kd disulphide-linked heterodimer comprised of one α chain (~50 000 mol. wt.) and one β chain (~40 000 mol. wt.) (Allison *et al.*, 1982; Haskins *et al.*, 1983; Meuer *et al.*, 1983). Recently, cDNA clones encoding one of the chains of the T-cell antigen receptor have been isolated from mouse (Hedrick *et al.*, 1984a) and human (Yanagi *et al.*, 1984) T-cell libraries. Sequence analysis of these cDNA clones demonstrates that they contain variable, joining and constant regions similar to those encoding immunoglobulins (Yanagi *et al.*, 1984; Hedrick *et al.*, 1984b). Furthermore, the cDNA clones hybridise to gene sequences capable of undergoing somatic rearrangements in T-cells (Hedrick *et al.*, 1984a). The mouse and human cDNA probes most probably detect the β gene family of the T-cell antigen receptor since partial protein sequence analysis of a human T-cell β chain agrees with the translated nucleotide sequence of the human cDNA clone (Acuto *et al.*, 1984). We have determined the chromosomal localisation of a human T-cell antigen receptor β chain gene using interspecific somatic cell hybrids and a mouse cDNA clone as a probe. The human gene maps to the distal portion of the long arm of chromosome 7.

Results

The mouse cDNA clone, 86T1 (Hedrick *et al.*, 1984b), was used to probe DNA isolated from human-mouse somatic cell hybrids. This clone contains sequences encoding the complete amino acid sequence of the mouse β chain together with a short segment of 5′-untranslated sequence, and was used without subcloning. The clone shares >80% overall sequence homology with a human clone (Yanagi *et al.*, 1984; Hedrick *et al.*, 1984b). It cross-hybridises with a 1.3-kb human T-cell mRNA species on Northern blot analysis and has been used to isolate an analogous clone from a cDNA library prepared from a human T cell line (M.K.L. Collins, M.J. Dunne, M.J. Owen and P.W.J. Rigby, unpublished observations).

A Southern blot of an *Eco*RI digest of human and mouse DNA revealed different size restriction fragments hybridising with the 86T1 probe (Figure 1). Two bands (2.7 kb and 11.7 kb) were observed with *Eco*RI-digested mouse L cell DNA (Figure 1, track A). With DNA prepared from a mouse T-cell line (BW5147) a 2.3-kb band was observed in addition to the 2.7-kb and 11.7-kb bands, reflecting the somatic rearrangement of the gene, from which the 86T1 probe is derived, in T-cell lymphomas (Haskins *et al.*, 1983) (Figure 1, track B). A Southern blot of human cell DNA, when probed at low stringency (2 x SSC) with the 86T1 insert showed two bands, at 11.7 kb and 4.7 kb (Figure 1, track C) with *Eco*RI. These bands correspond to a human T cell antigen receptor β chain gene since they showed somatic rearrangement when DNA from a human T cell lymphoma was used (Figure 1, track D).

The restriction fragment size differences between human

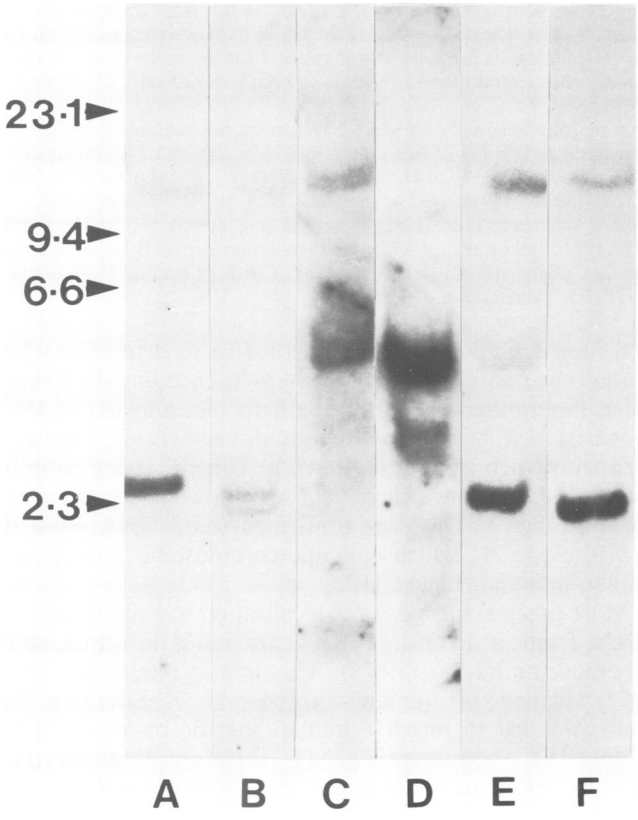

Fig. 1. Southern hybridisation analysis of human, mouse and somatic cell hybrid DNA. Samples were digested with *Eco*RI and electrophoresed and blotted as described in Materials and methods. Samples were: **(A)** IR, mouse L cell, **(B)** BW5147, mouse thymoma, **(C)** Molt-4, human thymoma, **(D)** JY, human B lymphoblastoid cell line, **(E)** clone 21E, **(F)** FIR5. Positions of λ *Hind*III markers are indicated.

Table I. Segregation of a human β chain gene in somatic cell hybrids

Hybrid	Human chromosomal contribution[a]	Presence of human-specific[b] bands
CTP41.P2[c]	2, 3, 6, 7, 14, 16, 17, 20, X	+
HORP27R C14[d]	4, 7, 10, 11, 12, 14, 15, 21	+
3W4 C15[e]	7, 10, 11, 12, 14, 15, 17, 21, X	+
2W1R70[e]	7, 13	+
Clone 21E[f]	7	+
DUR4.3[g]	3, 5, 10, 11, 12, 13, 14, 15, 17, 18, 20, 21, 22, X	−
DT1.2[h]	3, 8, 10, 11, 13, 15, 17, 18, 20, 21, 22, X	−
SIR74ii[i]	1, 2, 3, 12, 14, 18, 21, 22, X	−
FIR5[j]	7pter-1q22, 14, 18, Xqter-Xq13	−
Controls		
IR[e]	Mouse L cell	−
BW5147[k]	Mouse thymoma	−
PCC4[l]	Mouse embryonal carcinoma	−
Maja[m]	Human B-lymphoblastoid line	+
JY[m]	Human B-lymphoblastoid line	+
MOLT 4[n]	Human T-cell line	+
HSB2[o]	Human T-cell line	+

[a]Chromosomal presence or absence was determined by a combination of karyotypic, isozyme and antigenic analyses. In this table chromosomal fragments are indicated by scoring the whole chromosome positive except for rearrangements involving chromosome 7 for which the precise fragment is indicated. Several of the hybrids have been recloned since the original publication.
[b]Apart from human T cells which have undergone rearrangement the human diagnostic *Eco*RI band has a molecular size of 4.7 kb (see text).
[c]Jones *et al.* (1976); [d]Van Heyningen *et al.* (1975); [e]Nabholz *et al.* (1969); [f]Croce and Koprowski (1974); [g]Solomon *et al.* (1976); [h]Swallow *et al.* (1977); [i]Whitehead *et al.* (1982); [j]Solomon *et al.* (1983); [k]Hyman and Stallings (1974); [l]Jakob *et al.* (1973); [m]EB virus transformed B lymphoblastoid cell lines obtained from Dr. J. Bodmer (I.C.R.F., London); [n]Minowada *et al.* (1972); [o]Adams *et al.* (1968).

and mouse *Eco*RI-cut DNA enabled us to determine which human chromosome contained the gene encoding the β chain, using the mouse probe and Southern blot analysis of DNA prepared from human-mouse somatic cell hybrids. The use of a panel of such hybrids is shown in Table I. Using an initial panel of hybrids the β chain gene was shown to segregate with chromosome 7. This was confirmed using DNA from the hybrid clone 21, which contains chromosome 7 as its only human material (Figure 1E).

More precise chromosomal localisation was obtained using DNA from a hybrid, FIR5, containing a translocation chromosome t(x;7)(q13;q22). This hybrid retains the 7pter-7q22 segment of human chromosome 7. Southern blot analysis failed to reveal a human specific band with FIR5 (Figure 1F). The human β gene is, therefore, localised to the q22-qter region of the long arm of chromosome 7.

Discussion

These results demonstrate that a human β chain is localised on chromosome 7. This localisation excludes a linkage in the human system with the immunoglobulin heavy chain, kappa or lambda gene clusters (located on chromosomes 14, 2 and 22, respectively) (Croce *et al.*, 1979; Malcolm *et al.*, 1982; McBride *et al.*, 1982) or with the major histocompatibility complex (located on chromosome 6; Francke and Pellegrino, 1977), despite the striking similarities of the β chain genes with immunoglobulin heavy chain genes. This contrasts with a mouse T-cell antigen receptor β chain gene which has been localised to chromosome 6 (Lee *et al.*, 1984), which also carries the mouse kappa chain gene cluster (Hengartner *et al.*, 1977). The results presented here, however, would appear to rule out any functional importance for such a linkage since it has not been maintained in the human system.

Chromosomal translocations which bring the c-*myc* gene into the proximity of an Ig gene cluster are frequently observed in Burkitt's lymphoma (reviewed by Perry, 1983) and may participate in B-cell tumorigenesis. Similar translocations involving chromosome 7 are not generally associated with T-cell tumours, although deletion of the q22-qter region of the long arm or monosomy of chromosome 7 has been noted in a few leukaemias (de la Chapelle and Berger, 1984). These changes do not imply any obvious relationship between the T-cell receptor β gene and any known oncogene. The only chromosome 7 locus which is known to be related to an oncogene is the gene encoding the epidermal growth factor (EGF) receptor (Downward *et al.* 1984). Chromosomal localisation of the EGF receptor gene to 7p13-p11 (Meera Khan and Smith, 1984) places this locus a considerable distance away from the T-cell receptor gene. Further karyotypic and molecular analyses concentrating on chromosome 7 in T-cell leukaemias will be required to determine if a system analogous to the c-*myc* translocation is associated with T cell tumorigenesis.

Inspection of the human gene map does not indicate any immune function disorders or disease susceptibility loci associated with chromosome 7 (Meera Kahn and Smith, 1984). It might be predicted that this will be a fruitful area for further research.

Materials and methods

Cell lines and somatic cell hybrids

Cell lines and somatic cell hybrids used in this study are described in the references given in the footnote to Table I. Specific cell culture methods and conditions are also described in the same references. The human chromosomal content of somatic cell hybrids was determined by karyotypic analysis (Solomon *et al.*, 1976), isozyme analysis (Harris and Hopkinson, 1976) and antigenic analysis (Tunnacliffe and Goodfellow, 1984). The specific markers used for chromosome 7 were biliverdin reductase (Meera Khan *et al.*, 1983) and the EGF receptor which was assayed using the monoclonal antibody EGFR1 (Waterfield *et al.*, 1982).

Southern hybridisation analysis

High mol. wt. DNA was prepared from various human-mouse somatic cell hybrids and human and mouse cell lines as previously described (Maniatis *et al.*, 1982). 50 μg of DNA was digested to completion with *Eco*RI (purchased from Boehringer, 150 units, 16 h at 37°C). Following ethanol precipitation the digest was electrophoresed on an 0.8% agarose gel in Tris-acetate buffer at 100 mA. The DNA was partially depurinated and transferred to nitrocellulose after denaturation with alkali. The DNA hybridisation probe used was the purified 86T1 insert (Hedrick *et al.*, 1984b), which was nick-translated using [α-^{32}P]dCTP and [α-^{32}P]dATP to a specific activity of 1 x 10^8 d.p.m./μg. Hybridisation was performed at 68°C in 6 x SSC, 0.5% SDS, 1 mM EDTA, 5 x Denhardt's solution and 100 μg/ml salmon sperm DNA for 16 h. The filter was washed for 2 x 30 min in 2 x SSC, 0.1% SDS at 65°C, and exposed at −70°C using Fuji RX film and intensifying screens.

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Note added in proof

We have confirmed the results obtained with the mouse clone 86T1 using a human clone, isolated from a cDNA library from the human T-cell J6, as a probe. After this manuscript was accepted for publication Caccia *et al. (Cell,* **37,** 1091-1099) also mapped the human β chain of the T-cell antigen receptor to chromosome 7. Their results however localise this gene to bands p13-21. The reason for this discrepancy is unclear.