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Genetics

Murine *Ly-6* multigene family is located on chromosome 15

(chromosome mapping/somatic cell hybrids/restriction fragment length polymorphism/recombinant inbred lines/*in situ* hybridization)

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ABSTRACT **Murine *Ly-6*-encoded molecules play an important role in the antigen-independent activation of lymphocytes. We have described the cloning of a cDNA encoding the protein component of an Ly-6 molecule. Hybridization studies indicated that this cDNA identified multiple DNA fragments on Southern blots. The banding pattern exhibits a restriction fragment length polymorphism from mice bearing either the *Ly-6*^{a} or the *Ly-6*^{b} allele. We have employed three independent chromosomal mapping techniques, somatic cell hybrids, *in situ* hybridization, and strain distribution pattern analysis of the restriction fragment length polymorphism of DNA from recombinant inbred lines, to ascertain the chromosomal origins of these bands. We report that all members of the *Ly-6* multigene family are tightly linked on chromosome 15 and have been regionalized by *in situ* hybridization analysis to band 15E on the distal portion of this chromosome. Linkage analysis has indicated that the *Ly-6* genes are located within 1 map unit of *Env-54* (a retroviral envelope restriction fragment length polymorphism probe), 3 map units from *ins-1*, (insulin-related gene), and 4 map units from the protooncogene c-*sis*. The possible involvement of the *Ly-6* lymphocyte activation and differentiation antigen genes in chromosome 15-related lymphoid malignancies is discussed.**

The *Ly-6* genetic locus was first defined as controlling the expression of alloantigenic specificities on peripheral T lymphocytes (1). Ly-6-related specificities have since been described on several cell types, especially on B and T lymphocytes. Each of the cell-surface specificities controlled by the *Ly-6* locus appears to possess a distinct tissue distribution pattern (reviewed in refs. 2 and 3). Several reports suggest a role for *Ly-6*-encoded specificities in the process of antigen-independent lymphocyte activation (4–6). In a study of the thymic expression of the *Ly-6*-encoded specificity T-cell activating protein (TAP) (7), it was shown that TAP expression correlated with immunocompetence of cells in the thymic compartment and that TAP expression and function are not dependent on T-cell receptor expression (8). A separate study showed that cross-linking of the rat monoclonal antibody D7, which recognizes a nonpolymorphic determinant on *Ly-6*-encoded molecules, induced a potent T-cell proliferative response (6), suggesting that Ly-6 molecules may play a critical role in the T-cell activation cascade. The study of *Ly-6*-encoded molecules has been complicated by the presence of multiple specificities, each with an apparently distinct pattern of tissue expression.

In an attempt to better understand the complexities suggested by serological and biochemical analyses, we have initiated molecular genetic studies of the *Ly-6*-encoded molecules. Based on the amino acid sequence of one *Ly-6*-encoded protein, Ly6E.1 (9, 10), synthetic oligonucleotides were constructed and used to isolate an Ly6E.1 cDNA (11). The Ly6E.1 cDNA sequence contains information for a 26-amino acid leader peptide, followed by a cysteine-rich, 108-residue core protein with no N-linked glycosylation sites. The Ly-6 specificity TAP is anchored in the cell membrane by a phosphatidylinositol lipid linkage (12). The protein sequence deduced from the Ly6E.1 cDNA is similar to that reported for another lipid-linked membrane protein, Thy-1, in that the last 30 amino acids are predominantly hydrophobic, with no positively charged residues to define a transmembrane segment (11–14). Thus, although we have as yet no direct proof that the mature Ly6E.1 protein is attached to the cell membrane via a lipid linkage, the reported Ly6E.1 cDNA sequence is entirely consistent with such a notion. On Southern blots the Ly6E.1 cDNA hybridizes with multiple fragments of DNA from all strains and with every restriction enzyme tested exhibiting a restriction fragment length polymorphism (RFLP) that correlates with the *Ly-6* allele of the DNA donor. Preliminary results of genomic cloning studies suggest that at least 10 distinct *Ly-6*-related genes are identified by the Ly6E.1 cDNA (A.B., unpublished data).

The *Ly-6* gene complex was initially mapped by genetic techniques to chromosome 9 (15) and was later reassigned to chromosome 2 (16). This paper reports the use of the Ly6E.1 cDNA to establish the chromosomal location of the *Ly-6* genes by DNA hybridization methods. The location was determined by Southern blot analysis of DNA from a panel of hamster–mouse somatic cell hybrids, by *in situ* chromosomal hybridization, and by analysis of the hybridization pattern of DNA from a large number of recombinant inbred mouse lines.

METHODS

Somatic Cell Hybrids. The somatic cell hybrids used for the present assignment were produced by fusion of the Chinese hamster cell line E36 with embryo fibroblasts, peritoneal macrophages, sarcoma cells, or established cell lines from the following mouse strains: BALB/c (hybrid clones mFE2/1/1, mFE2/1/7, TuCE12/G1, TuCE12G/2, TuCE12G/4, TuCE12/G7, mal0b, CEC), C3H (R44, Ecm4e), A/HeJ (4B31Az3, 2aC2), or noninbred CD-1 mice (C11, C17B). The inbred strains from which the mouse parental strains were derived are known to carry the *Ly-6*^{a} allele. The chromosome composition of the hybrids was determined by trypsin–Giemsa banding (17, 18) and by isozyme analysis (19).

Southern Blotting. For Southern blot analysis, DNA from the parental and somatic cell hybrid lines and from isolated liver nuclei of the A × B and B × A recombinant inbred (RI) strains was isolated by standard procedures. DNA from non-RI lines was purchased from DNA resources of The

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Abbreviations: RFLP, restriction fragment length polymorphism; RI, recombinant inbred; SDP, strain distribution pattern; TAP, T-cell activating protein.

Jackson Laboratory. For Southern blotting, DNAs were digested with *Eco*RI then loaded directly onto 0.8% agarose gels and processed as described (11, 20). The 750-base-pair Ly6E.1 cDNA insert of pKLy6E.1-2R, which contains 75 base pairs of 5′-untranslated region, the entire coding region, and ≈380 base pairs of the 3′-untranslated region, was nick-translated to a specific activity of at least 6.2×10^8 cpm/μg, hybridized to, and washed from the filters as described (11).

***In Situ* Hybridization.** Metaphase chromosome spreads were prepared from primary mouse embryo cell cultures as described by Disteche *et al.* (21). Chromosomes were G-banded and photographed, after which they were destained and stored dessicated at −20°C until used for *in situ* hybridization. The Ly6E.1 cDNA probe was nick-translated with ^{125}I-labeled dCTP (Amersham; >1500 Ci/mmol; 1 Ci = 37 GBq) to a specific activity of 6×10^8 dpm/μg and hybridized to the chromosome preparations using published procedures (22). Slides were autoradiographed for 14 days.

Mapping Using RI Strains. The A × B/Ns and B × A/Ns recombinant inbred strains were developed at the University of California at San Diego from the inbred strains A/J and C57BL/6J (23). The A × B and B × A RI strains are currently distributed between F_{21} and F_{50} of inbreeding. Sufficient segregation still occurs in strains below F_{30} to create a real danger of mistyping a strain when a determination is done on DNA from a single individual. We, therefore, typed DNA from both members of the "in-line pair" for strains not yet at F_{30}. Single individuals were typed for strains at or beyond F_{30}. Using RI strains to determine the existence and strengths of linkage between two loci involves simply asking how often strains inherit their alleles at the two loci from different progenitor strains. If the two loci are very close together, their alleles will almost always come from the same progenitor. Taylor (24) has described the relationship between the distance between the two loci and the freqency with which they recombine (i.e., are not inherited from the same progenitor strain) in RI strains. The relationship is described by the equation, $r = R/(4-6R)$, where r is the recombination fraction (recombination fraction times 100 is the map distance in centimorgans) between the two loci, and R is the fraction of recombinants in a set of RI strains. Silver (25) described how confidence intervals should be calculated for estimates of linkage distances derived from RI strains. A BASIC computer program called LINKAGE was written (by M.N.N.) that compares pairs of strain distribution patterns, identifies instances of recombination, and calculates map distances and confidence intervals by the methods of Taylor (24) and Silver (25). This program was used for the calculations presented in this paper. The strain distribution patterns (SDPs) for the chromosome 15 markers have been determined by M.N.N. (unpublished data).

RESULTS

Somatic Cell Hybrid Mapping of the *Ly-6* Genes. Our initial approach to determining the chromosomal location of the *Ly-6* multigene family involved a study of DNA hybridization of the Ly6E.1 cDNA probe to DNA from a panel of hamster–mouse somatic cell hybrids. These results showed a clear difference in the hybridization pattern of mouse DNA (≈15 bands) and hamster DNA (one major cross-reacting band), indicating that it would be possible to discriminate mouse-derived DNA hybridization from the hamster DNA background in the somatic cell hybrids under the washing stringency conditions used (2× SSC, 65°C; 1× SSC = 0.15 M NaCl/0.015 M sodium citrate, pH 7.0.). A determination of the hybridization profile (mouse or hamster) and the list of mouse chromosomes retained by each of the hybrids, as determined by karyotype and isozyme analysis, are presented in Fig. 1. The hybridization results indicated that either all, or none, of the mouse-derived hybridizing bands were present in any one somatic cell hybrid, which suggests that the hybridizing DNA is derived from a single mouse chromosome. Correlation of the hybridization pattern with the presence of specific mouse chromosomes indicates a 0% discordance only for chromosome 15. The results from

Gel Lane	DNA	Hybridization	MOUSE CHROMOSOME 1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	X
1	CMS4625	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
2	E36	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−
3	mFE2/1/7	+	+	+	+	−	−	+	+	+	+	−	−	+	+	−	+	−	+	−	+	+
4	mFE2/1/1	+	+	+	+	+	−	+	+	+	+	+	−	+	+	−	+	−	+	+	+	+
5	2a C2	+	+	+	+	+	−	−	+	+	+	+	−	+	+	−	+	+	+	+	+	+
6	TuCE12G/1	−	−	−	−	−	+	*	−	*	−	−	−	+	+	+	−	+	+	−	+	+
7	TuCE12G/7	+	−	+	−	−	+	*	−	*	−	+	−	+	+	+	+	+	+	+	+	+
8	C 11	+	+	+	+	+	+	+	+	+	+	+	−	+	+	−	+	+	+	−	+	+
9	C 17/B	+	+	+	+	+	−	−	+	−	+	−	−	+	−	−	+	−	−	−	+	*
10	4B31Az3	+	−	+	−	−	−	−	+	−	−	−	−	+	−	−	+	+	−	−	+	−
11	R 44	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	−	+	+	−	−
12	Ecm4e	+	−	−	−	−	−	−	−	−	−	−	−	−	−	*	+	−	−	−	−	−
13	ma 10b	−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	−	−	−	−	−	+
14	CEC	−	−	−	−	−	−	*	−	−	−	−	−	−	−	−	−	−	−	−	−	+
15	TuCE12G/2	+	−	−	−	−	−	*	−	*	−	−	−	−	+	−	+	+	+	−	−	+
16	TuCE12G/4	+	−	−	−	−	+	*	−	*	−	+	−	+	+	+	+	+	+	+	+	+
	Percent Discordance		36	21	36	43	57	44	29	30	31	36	71	29	29	62	0	36	36	50	21	38

FIG. 1. Mapping of the *Ly-6* gene family using somatic cell hybrids. A listing of the origin of the DNA in lanes 1–16 is shown with a determination of whether the hybridization exhibits the background hamster (−) or *Ly-6*-related (+) profile (boxed symbols). The karyotype analysis of the individual somatic cell hybrids is shown indicating the deletion (−) or the retention of a normal (+) or of a rearranged (*) mouse chromosome. An analysis of the hybridization profile and the presence or absence of each mouse chromosome is shown at the bottom as percent discordance. Rearranged chromosomes are excluded from this analysis.

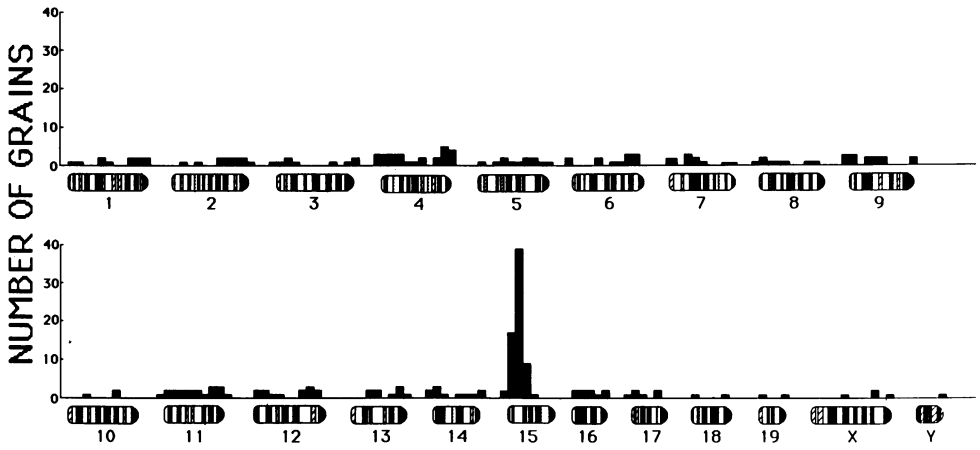

FIG. 2. *In situ* hybridization mapping of the *Ly-6* genes. The murine *Ly-6* gene cluster was localized to chromosome 15 by *in situ* hybridization of ^{125}I-labeled Ly6E.1 cDNA probe to chromosome spreads prepared from primary mouse embryo fibroblasts. In this figure the mouse karyotype idiograms are oriented with the centromeres to the right.

specific hybrids rule out chromosome 2 (lanes 12, 15, and 16) as well as chromosome 9 (lanes 7, 10, 12, 15, and 16) as the location for the *Ly-6* genes as has been reported (15, 16).

***In Situ* Hybridization Mapping of the *Ly-6* Genes.** To confirm the assignment of the *Ly-6* genes to chromosome 15, and to determine their position along the chromosome, *in situ* hybridization was performed. The distribution of autoradiographic labeling observed over chromosomes after hybridization with the ^{125}I-labeled Ly6E.1 cDNA was plotted on a computer-generated histogram in which a standard idiogram of the mouse karyotype (18) was divided into 156 units proportional to an average silver grain diameter of 0.35 μm. A total of 32 metaphase spreads were analyzed. Background labeling was low, averaging less than 2 grains per unit chromosome length. Of 262 silver grains associated with chromosomes, 68 (26%) were located on chromosome 15 as shown in Fig. 2. A total of 65 (96%) of the chromosome 15-specific grains were concentrated within the region D3-F, with a peak over band E, the most probable locus of the *Ly-6* gene cluster. This region comprises $\approx$0.6% of the haploid murine genome. Statistical evaluation by Poisson distribution of the number of silver grains within the histogram peak indicated that these data were highly significant ($P < 0.001$).

Mapping by RI Line RFLP Strain Distribution Pattern Analysis. Recombinant inbred line analysis was employed as another independent method for confirming the chromosomal location of the *Ly-6* genes and to position the *Ly-6* genes in relation to other genetic markers on chromosome 15. A requirement for using RI lines in mapping is that the progenitor strains differ in some detectable way for the gene of interest. We have defined a RFLP between the *Ly-6*^{a} and the *Ly-6*^{b} alleles in all strains tested, including the C3H and its *Ly-6* congenic, C3H.B6-Ly-6^b. We have found an absolute correlation between the RFLP pattern and the known *Ly-6* allele for the 7-member BALB/c × C57BL/6 (CXB), and the 12-member C57BL/6 × C3H (BXH) RI line panels (ref. 11 and K.P.L., unpublished data). Since larger panels lead to better linkage data, we have also made use of the 50 members of the A × B and B × A RI strain panels developed by Nesbitt and Skamene (23). The strain distribution patterns for over 150 markers have been determined for this RI panel. Although the individual A × B and B × A RI lines have not been serologically typed for *Ly-6* allele expression (*Ly-6*^{a} or *Ly-6*^{b}), we feel that the perfect correlation observed to date (52 strains tested) between RFLP and *Ly-6* allele, warrants their use for this purpose. The RFLP pattern obtained after probing DNA from each member of the A × B and B × A RI line panel is listed in Table 1. This strain distribution pattern was compared with the SDP of other markers. Significant evidence for linkage between *Ly-6* and other markers was seen only with markers on chromosome 15. Table 1 shows the established SDPs of these relevant markers compared to the *Ly-6* SDP defined here. Table 2 shows the matrix of linkage distances among the chromosome 15 markers. These distance estimates do not yield a unique solution in terms of gene order. However, the most likely order, chosen so as to fit the linkage distance estimates and to minimize the number of postulated double and triple crossovers is either

Ag-1–Ly-6–Env-54–c-sis–Ins-1–Ker-1–Int-1–Gdc-1

or

Ag-1–Env-54–c-sis–Ly-6–Ins-1–Ker-1–Int-1–Gdc-1.

DISCUSSION

Using three independent chromosome mapping procedures, we have assigned the murine *Ly-6* genes to chromosome 15. The results from the somatic cell hybrid mapping suggested that all of the *Ly-6*-related genes detected by our Ly6E.1

Table 1. SDPs of chromosome 15 markers

Marker	SDP									
	(A × B) 1–25					(B × A) 1–25				
Ag-1	abbba	aabba	baobb	obaoa	booob	obbbo	ooooo	bboao	boooo	ooboo
Env-54	aabbb	abbob	baobb	obaoo	boaob	bbbba	bbabo	bbbao	babbo	obbbo
c-*sis*	aabba	abbab	babba	obaaa	boaab	bbbbo	baabb	bbbao	babbb	obbbb
Ly-6	**aabbb**	**abbab**	**babbb**	**oobbb**	**bbaab**	**bbobo**	**bbabb**	**bbbao**	**babbb**	**obbbb**
Ins-1	aaobo	abbob	baoba	ooaoo	aoaoo	bobba	boaoo	bbbao	baooo	obboo
Ker-1	abbba	bbaab	baoaa	oaaoa	aobob	bobaa	obooo	babao	bobao	oaboo
Int-1	abbba	bbobb	oaoao	oaaaa	aobbo	oobaa	ooooo	baoao	babao	oaboo
Gdc-1	abbbo	bbaab	oaaao	oaaoa	aobba	aobaa	booao	baboo	babao	oaboo

The 50 columns across the table represent the RI strains A × B 1 through A × B 25 (first 25 columns) and B × A 1 through B × A 25 (columns 26–50). The letters a and b represent the A/J-derived and the C57BL/6-derived alleles, respectively. Where an o appears it indicates that the strain was not typed or that it was still segregating. The *Ly-6* SDP is shown in bold letters.

Table 2. Matrix of linkage distances between chromosome 15 markers

Marker	Chromosome 15 linkage matrix						
	Ag-1	*Env-54*	c-*sis*	*Ly-6*	*Ins-1*	*Ker-1*	*Int-1*
Env-54	0.05*	—	—	—	—	—	—
c-*sis*	0.06*	0.02*	—	—	—	—	—
Ly-6	**0.11***	**0.01***	**0.04***	—	—	—	—
Ins-1	0.10*	0.02*	0.01*	**0.03***	—	—	—
Ker-1	0.29	0.26	0.20	**0.39**	0.15	—	—
Int-1	0.20	0.29	0.27	**0.50**	0.17	0.01*	—
Gdc-1	0.50	0.39	0.44	**0.50**	0.21	0.02*	0.01*

Numbers represent the linkage distances expressed as the recombination fraction between two chromosome 15 markers, where * indicates a linkage significant at the 95% confidence level. Values for *Ly-6* are in bold letters.

cDNA probe are contained on this single chromosome. This was confirmed by the data from the *in situ* mapping, which also indicated that the *Ly-6* genes are clustered in the region of band E on the distal end of chromosome 15. These results are consistent with immunogenetic studies that used backcross analysis to establish that the *Ly-6* genes are genetically tightly linked. Pulsed-field gradient electrophoretic analysis and genomic cloning of the genes in the *Ly-6* locus should provide estimates of actual physical linkages among the members of the *Ly-6* gene family.

Our assignment of the *Ly-6* genes to chromosome 15 is in conflict with the reported chromosomal assignment(s) for this locus (15, 16). Using anti-Ly-6.2 sera, Horton and Hetherington (15) demonstrated close linkage (15.7 ± 2.1 recombination units) between the *Ly-6* and *Thy-1* loci in several backcross combinations and were led to position *Ly-6* on chromosome 9. This assignment was later challenged by Meruelo *et al.* (16) who used anti-Ly-6.2 monoclonal antibodies and were unable to confirm the *Thy-1–Ly-6* linkage. They postulated that the results obtained by Horton and Hetherington (15) may have been attributable to complexities in the sera used.

Meruelo *et al.* (16) used linkage relationships to known markers to position the gene for the lymphocyte specificity Ly-11 to mouse chromosome 2. They had shown an identical strain distribution pattern in the 7-member CXB RI lines between *Ly-6* and *Ly-11* (26). Segregation analyses had also shown a concordance in *Ly-6* and *Ly-11* genotype in 46 of 50 progeny of crosses between (A/J × B10)F_1 mice (26). Based on this apparent linkage between *Ly-6* and *Ly-11*, they mapped *Ly-6* to a position on chromosome 2. Evidence was also presented for a linkage of *Ly-6* to a locus influencing susceptibility to radiation-induced leukemia, *Ril-1* (27), and to the minor histocompatibility locus *H-30*, which were both assigned to chromosome 2 (16). Further examinations of this and other work has led to a reevaluation of these chromosomal assignments. It is now believed that *Ly-11* does in fact map to chromosome 2, but that the other markers, *Ly-6*, *Ril-1*, and *H-30*, should be reassigned to mouse chromosome 15 (D. Meruelo, personal communication).

Although each of the three chromosome mapping procedures employed in this study mapped the *Ly-6*-related genes to mouse chromosome 15, all involved the hybridization of the Ly6E.1 cDNA to chromosomal DNA. Lacking a cDNA, both previous *Ly-6* mapping studies relied on serologically-assayed cell surface expression of Ly-6 specificities. Although the Ly6E.1 protein does not appear to contain N-linked carbohydrate (9), at least five charged species with pI values between 4 and 5.2 have been identified (10), suggesting extensive post-translational modifications before cell-surface expression of the Ly6E.1 specificity. The Thy-1 protein, another lymphocyte cell surface specificity that has been implicated in cellular activation (28), requires extensive post-translational processing, including lipidation (13, 14). Several classes of Thy-1$^-$ mutant cell lines have been described (29). Interestingly, two mutant classes have been shown to also be deficient in the expression of certain Ly-6 specificities (30). These findings suggest that common lipidation and other post-translational pathways may be involved in the expression of Thy-1 and Ly-6 specificities, which may explain some of the difficulties encountered by others, and especially Horton and Hetherington (15), in their attempts to map the *Ly-6* genes based on cell-surface expression studies.

Ly-6-related genes have been shown by several groups to play a critical role in the process of lymphoid cell activation (4–6). The assignment of the genes of the *Ly-6* multigene family to chromosome 15 prompted an analysis of published reports concerning chromosome 15, with special considerations given to lymphoid malignancies, as reviewed by Klein (31). In a study of the spontaneous thymomas of AKR mice, Dofuku *et al.* (32) reported a frequent association with trisomy of chromosome 15. A similar predominance of chromosome 15 trisomy was observed in T-cell leukemias induced in C57BL mice by exposure to radiation leukemia virus or to the chemical carcinogen dimethylbenz[*a*]anthracene (33–35). The duplication of the distal portion of chromosome 15 was frequently associated with the occurrence of T-cell leukemias (36, 37). As mentioned above, the major locus influencing susceptibility to leukemia induction by fractionated irradiation, *Ril-1*, has been remapped to a position on chromosome 15, closely linked to the *Ly-6* locus (27, 38). As indicated in Fig. 3, the murine leukemia virus integration site *Mis-1* and the Moloney murine leukemia virus integration sites *Mlvi-1* and *Mlvi-2* have been assigned to chromosome 15. The SDP data presented here (Table 1) indicates that the *Ly-6* genes are tightly linked to the marker *Env-54* (Table 2). *Env-54* is defined by an RFLP observed between A/J and C57BL/6J mice using an *env* gene fragment of the Moloney mink-cell focus-inducing (MCF) virus as a probe (39–41).

In addition to the chromosome 15 trisomy frequently observed in T-cell lymphomas, numerous translocations of chromosome 15 involving the c-*myc* gene have been associated with B-cell plasmacytomas. The c-*myc* gene has been mapped to the vicinity of bands 15 D2-3 by translocation breakpoint analysis (42, 43) and by *in situ* hybridization (44). As c-*myc* is not one of the markers contained in the A × B and B × A RI panel and as *in situ* hybridization provides only low resolution assignments, we have as yet no direct estimates of the genetic or physical linkages between c-*myc* and the *Ly-6*-related genes. Although the majority of plasmacytomas characterized to date exhibit translocations directly involving the c-*myc* gene, 10–25% of these tumors have variant translocations. A chromosome 15 breakpoint found in several plasmacytomas, called the plasmacytoma variant translocation (*pvt-1*) locus located at least 72 kilobases distal to c-*myc* has been described (45, 46). The DNA of the *pvt-1* locus showed no homology to the chromosome

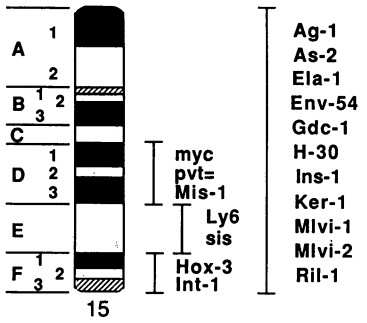

FIG. 3. Gene map of mouse chromosome 15. Markers that have been assigned to a specific chromosomal region by physical means are indicated by the narrow brackets. Other markers that are contained on chromosome 15 but have not been specifically localized, are presented alphabetically at the right. Markers are as follows: *Ag-1*, Agouron Institute DNA polymorphism-1; *As-2*, aryl sulfatase; *Ela-1*, elastase; *Env-54*, retroviral envelope RFLP-54; *Gdc-1*, glycerol 3-phosphate dehydrogenase; *H-30*, histocompatibility locus-30; *Hox-3*, homeo-box locus-3; *Ins-1*, insulin-related sequence; *Int-1*, mouse mammary tumor virus integration site; *Ker-1*, keratin; *Mis-1*, murine leukemia virus integration site; *Mlvi-1,2*, Moloney murine leukemia virus integration sites; *myc*, myelocytomatosis viral oncogene homologue; *pvt-1*, plasmacytoma-associated variant translocation locus; *Ril-1*, radiation-induced leukemia susceptibility locus-1; *sis*, simian sarcoma virus oncogene homologue.

15 genes c-*sis* (v-*sis*), *int-1*, the putative mammary oncogene, or to any of 16 known oncogenes (45). Several T lymphomas were described that contain mink-cell focus-inducing proviral inserts in the *pvt-1* region (47). It was subsequently shown that *pvt-1* is equivalent to *Mis-1*, the murine leukemia virus integration site mentioned above (48). The relationship, if any, between the DNA of the *pvt-1/Mis-1* region and the *Ly-6*-related genes and/or their tightly linked mink-cell focus-inducing viral marker, *Env-54*, should be determined.

This study assigns the *Ly-6* family of genes, encoding lymphoid differentiation antigens involved in cellular activation, to the vicinity of band E on mouse chromosome 15. This portion of chromosome 15 contains the protooncogenes c-*sis* and c-*myc*, both involved in the regulation of cell growth. The observations implicating this segment of chromosome 15 in both B- and T-cell malignancies have been discussed and suggest further studies of the oncogenic potential of *Ly-6*-related genes.

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