RESISTANCE GENES TO MURINE LEUKEMIA IN THE I IMMUNE RESPONSE GENE REGION OF THE H-2 COMPLEX

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It is generally believed that one of the elements of host resistance to leukemia is the immune response. Immune responsiveness to many antigens is under genetic control, regulated by Ir genes linked to the major histocompatibility complex (1). Ir gene regulation in the mouse is most likely based on two complementing Ir genes localized within the H-2 complex (2).

Involvement of the H-2 complex in resistance to leukemogenesis is well established (3). An H-2-linked resistance gene against leukemogenesis by Gross leukemia virus, Rgv-1, was described and localized by Lilly (4). The genetic position of Rgv-1 is to the left of the right border of the H-2 complex (H-2D), either within H-2 or to the left of it (4). Evidence for involvement of a locus (X-1) with similar localization to Rgv-1, in immune responsiveness to transplantable, radiation-induced leukemias was presented by Sato et al. (5). These findings suggest that H-2-linked Ir genes are involved in the host's immune response to leukemogenesis. More recently Chesebro et al. have described a resistance gene for Friend virus leukemogenesis, RFV-1 and localized it to H-2D (6). It has been shown that the H-2 complex contains a large number of loci, which control several different immunological functions, such as complement components and target antigens for alloimmune killer cells. Thus the distinctly localized Ir genes are only a part of the H-2-linked loci. In light of these data, the genetic localization of loci Rgv-1 and X-1 is not entirely sufficient for their identification with Ir genes, because of the limited genetic material investigated at the time of their description (4, 5).

In this report we present evidence for the localization of a resistance gene to virus-induced murine leukemia within the left part of the I region, and suggest that it is in complementation with an additional gene also situated within the H-2 complex. This genetic localization is consistent with that of the complementing Ir genes (2).

Materials and Methods

Animals. All mouse strains used in this study were bred in our Animal House. The breeding nuclei were either gifts from Doctors D. C. Shreffler (Washington University, St. Louis, Mo.), H. O. McDevitt (Stanford University, Stanford, Calif.), F. Lilly (Albert Einstein Medical Center, Bronx, N. Y.), or purchased from The Jackson Laboratories (Bar Harbor, Maine).

Virus Preparation. A-RadLV, a line highly oncogenic in young adult C57BL/6 mice, originated in the bone marrow of irradiated mice, and is passaged in vivo as a cell-free supernate of homogenates from leukemic thymuses, as described previously (7).
Treatment and Diagnosis.

0.02 ml of a 20% (on the basis of the weight of the thymus tissue) cell-free supernate was injected into the thymus of young adult mice. After approximately 2 mo the animals were inspected weekly and the sick mice were isolated. The animals were sacrificed in a terminal state and autopsied, and when necessary histological examination was performed. The genetic evaluation of the results was based on works by Shreffler and David (8), Klein (9), David (10), and Murphy et al. (11).

Results and Discussion

For the first experiment mouse strains were selected on the basis of their H-2 and Fv-1 type. Where the availability of congenic strains permitted, the same independent H-2 haplotype was tested on at least two different backgrounds.

Two conclusions could be drawn from this experiment (Table I). First it became apparent that the Fv-1^b type congenic series on the B6, B10, BALB, and A backgrounds are sensitive, while the Fv-1^a type on the C3H and DBA mice are resistant to leukemogenesis by a variant of the radiation leukemia virus (A-RadLV). This observation is in agreement with the known effect of the Fv-1 resistance locus (3), and with the in vitro studies of Declève et al. which suggest that RadLV is an Fv-1 B-tropic virus (12). The second conclusion was that the leukemogenic effect of A-RadLV depends on the H-2 haplotype in Fv-1 ~ mouse strains. It can be seen from Table I that haplotypes r, b, k, and d (in this order) are sensitive, f is probably intermediate, while s is resistant to leukemogenesis by A-RadLV.

In the subsequent experiments strains carrying chromosomal recombinations within the H-2 complex were investigated. These strains were selected either for recombination between the resistant s and the sensitive k and d haplotypes, or for recombination between two sensitive haplotypes like k and b, d and b, or b and a. The first group could give information on the localization of a resistance locus within the H-2 complex on the basis of the suggested position of the recombination event in these strains. The second group was planned to detect whether some of these sensitive genotypes would complement for resistance. Additionally haplotypes y1 and y2 were tested, and were found to be sensitive to leukemogenesis with A-RadLV, suggesting that q may be an additional sensitive H-2 haplotype. The two main groups (t2, t3, t1, an1 and h4, g, a, i, i5, respectively) however gave more detailed information which will be analyzed separately in Tables III and IV.

It can be seen in Tables II and III, that haplotypes t1 and an1 (strains A.TL and A.TFR1) which contain elements of the resistant s and of the sensitive k, a, or f haplotypes were highly sensitive, while t2 and t3 which are similar to these in origin, but differ in the position of the recombination, were resistant to leukemogenesis by A-RadLV. These data suggest that a resistance gene is localized between H-2K and subregion I-E in the left part of the I region of the H-2 complex. The left limit of this localization is indicated by the sensitivity of strains A.TL and A.TFR1 which carry the H-2K^a allele of the resistant A.SW strain and have sensitive k alleles in the I region. The right limit is indicated by resistance in the B10.HTT strain which have resistant s alleles in H-2K and in subregions I-A, I-B, and I-J, and sensitive k and d alleles in the rest of its H-2 complex. Additionally, Tla types a, c, and b appear in this group, both in the resistant and in the sensitive strains, suggesting that this locus is not involved in the control of leukemogenesis by A-RadLV in the tested genotypes. We
conclude from these data that in mouse strains carrying the permissive $Fv$-1 allele, viral leukemogenesis is most likely under the control of a locus within the left part of the $I$ region. We propose $Rrv-1$ (resistance to radiation leukemia virus) as the preliminary designation for this locus. It is noteworthy that the localization of $Rrv-1$ is very similar to that of most known $Ir$ genes (1, 8).

Another interesting observation was made in recombinant strains between sensitive haplotypes. Five such recombinant haplotypes were tested, $g$, $h^4$, $i$, and $i^5$. It can be seen from Tables II and IV that one of these five strains, B10.A(5R), despite being the descendent of the sensitive $b$ and $a$ haplotypes, was resistant to A-RadLV. The most plausible explanation for phenotypic resistance in a genotype which derives from the recombination of loci from sensitive origins, is the complementation of two resistance genes. In line with this explanation the data analyzed in Table IV, suggest that in the origin of recomb-
Localization of a Resistance Locus, Rrv-1, for Leukemogenesis by A-RadLV

### Table III

| H-2    | Origin | H-2K | I-A | I-B | I-J | I-E | I-C | S | H-3G | H-2D | Tin | Leukemia |
|--------|--------|------|-----|-----|-----|-----|-----|---|------|------|-----|---------|
| CS7B6/10M | b      | b    | b   | b   | b   | b   | b   | b | b    | b    | 100 | 11      |
| B10.BR/Sg  | k      | k    | k   | k   | k   | k   | k   | k | k    | k    | 100 | 11      |
| B10.D2/nSsn | d      | d    | d   | d   | d   | d   | d   | d | d    | d    | 77  | 11      |
| B10.S5/Sg  | s      | s    | s   | s   | s   | s   | s   | s | s    | s    | 11  | 1      |
| A.TL/Sf    | d2     | s/a  | s   | s   | s   | s   | s   | s | s    | s    | 12  | 1      |
| B10.H7T/Ph | d3     | stl  | s   | s   | s   | s   | s   | s | k    | k    | 13  | 1      |
| A.TL/Sf    | d1     | s/al | s   | k   | k   | k   | k   | k | k    | k    | 86  | 1      |
| A.TFR1/Sf  | d4/l   | s/a  | s   | k   | k   | k   | k   | k | k    | k    | 96  | 1      |

**Notes:**
- The localization of the second locus is indicated by the sensitivity of another b/a recombinant, HTI, and suggests that this locus is situated to the right of H-2D and to the left of Rrv-1. The complementation of two loci inside the H-2 complex is a characteristic of Ir genes. In one of the best known examples, for the immune responsiveness to the synthetic antigen L-Glu,L-Lys,L-Phe, Dorf et al. (2) have denoted the two loci as α and β. There is a striking similarity between their genetic data on the localization of complementing Ir genes for the regulation of an antibody response, and ours on the regulation of sensitivity to leukemogenesis. The data presented here strongly support the view that one of the functions of the major histocompatibility complex in leukemia may be analogous to that of Ir genes.

### Table IV

**Complementation of Resistance Genes for Leukemogenesis by A-RadLV within the H-2 Complex**

| H-2    | Origin | H-2K | I-A | I-B | I-J | I-E | I-C | S | H-3G | H-2D | Tin | Leukemia |
|--------|--------|------|-----|-----|-----|-----|-----|---|------|------|-----|---------|
| B10.A(4R)/Sg | h/4   | d/b | b   | b   | b   | b   | b   | b | b    | b    | 100 | 11      |
| B10.H7T/Ph  | g      | d/b | d   | d   | d   | d   | d   | d | b    | b    | 68  | 11      |
| B10.A/Sg    | a      | d/b | k   | k   | k   | k   | k   | k | d    | d    | 97  | 11      |
| HTI/G6     | i      | b/a | b   | b   | b   | b   | b   | b | b    | b    | 88  | 11      |
| B10.A(5R)/Sg| d/s   | b/a | b   | b   | b   | k   | k   | k | d    | d    | 11  | 11      |

**Notes:**
- The resistant vs. sensitive H-2 haplotypes for A-RadLV leukemia were found to be different from those for the very similar Gross virus leukemia (3, 4). This may indicate that there is a degree of polymorphism in the H-2-linked regulation of sensitivity to murine leukemia. Therefore, it will be of special interest to study the effect of the H-2 complex on leukemogenesis by the closely related original RadLV described by Kaplan (13).

**Summary**

A resistance locus to leukemogenesis in mice by A-RadLV (a variant of the
radiation leukemia virus) is described. This locus, Rrv-1, was mapped to subregions I-A, I-B, and I-J of the H-2 complex. It is suggested that Rrv-1 may be in complementation with a second locus to the right of it, between Rrv-1 and H-2D. This localization and the complementation of the two loci for resistance are characteristics similar to Ir genes, and indicate a possible relationship between the genetic regulation of immune responsiveness and susceptibility to leukemia.

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