Consequences of Self and Foreign Superantigen Interaction with Specific V_β Elements of the Murine TCR$\alpha\beta$

J.W. KAPPLER,†‡ A. PULLEN,* J. CALLAHAN,§ Y. CHOI,* A. HERMAN,*
J. WHITE,* W. POTTS,‖ E. WAKELAND,‖ AND P. MARRACK*†‡§

Howard Hughes Medical Institute, Division of Basic Immunology, Department of Medicine, National Jewish Center for Immunology and Respiratory Medicine, Denver, Colorado 80206; Departments of †Medicine, ‡Microbiology and Immunology, and §Biophysics, Biochemistry, and Genetics, University of Colorado Medical School, Denver, Colorado 80220; ‖Department of Pathology, University of Florida College of Medicine, Gainesville, Florida 32610

The $\alpha\beta$ T-cell receptor (TCR$\alpha\beta$) recognizes a ligand composed of an antigen fragment complexed with a product of the major histocompatibility complex (MHC). The repertoire of receptors is limited both by the germ line of receptor variable elements and by selective events that take place during T-cell development. The current view is that the germ-line repertoire is expressed in the thymus randomly but that only those T cells bearing receptors that successfully interact with MHC molecules expressed on thymic cortical epithelial cells are allowed to mature (Bevan and Fink 1978; Zinkernagel et al. 1978; Kisielow et al. 1988; Sha et al. 1988). Furthermore, during the process of establishing tolerance to self-antigens, this positively selected population is further reduced by the deletion or inactivation of clones whose receptors continue to interact with self-antigen/MHC ligands (Kappler et al. 1987a, 1988; MacDonald et al. 1988; Pullen et al. 1988). Thus, positive and negative selections reduce expressed receptor repertoire in the periphery to a fraction of the germ-line repertoire.

Five variable elements contribute to the specificity of the TCR$\alpha\beta$: V_α (variable), J_α (joining), V_β, D_β (diversity), and J_β. Despite the fact that in most circumstances all five components together determine receptor specificity, a study of the variation of expression of individual V_β elements in different mouse strains has produced a wealth of information on genetic factors effecting the TCR$\alpha\beta$ repertoire. Several studies have shown the influence of thymic-positive selection on expression of particular V_β elements (Blackman et al. 1989; Palmer et al., this volume). More striking are the many mechanisms in mice that prevent the expression of particular V_β elements in individual mice. Some of these are genetic defects within the V_β complex (Chou et al. 1987a; Kappler et al. 1987b; Wade et al. 1988). However, the most surprising mechanism limiting V_β expression involves the induction of tolerance to a set of unusual self-antigens, which we have termed "superantigens" (White et al. 1989). Superantigens combine with MHC molecules to generate ligands that are recognized by all TCR$\alpha\beta$s bearing one or a few V_β elements with little input from the other receptor variable elements (Kappler et al. 1987a, 1988; MacDonald et al. 1988; Pullen et al. 1988). The consequence is that mice carrying these antigens delete nearly all T cells bearing the relevant V_β elements while establishing self-tolerance. Since there are only a total of 22 different V_β elements in the mouse, these self-superantigens drastically alter the repertoire of the mice in which they occur.

Superantigens have also been identified as products of microorganisms. Particularly striking examples are the toxins of *Staphylococcus aureus*, which cause a number of diseases in humans and other animals (Bergdoll 1983). These toxins have been recognized as powerful T-cell stimulants for many years, but the role of T cells in their mode of action has not been pursued. Recently, we and other investigators have shown that these toxins are superantigens in that each stimulates a set of human or mouse T cells bearing particular V_β elements (Choi et al. 1989; Janeway et al. 1989; Kappler et al. 1989b; White et al. 1989).

In this paper, we review some of the properties of the bacterial and the polymorphic murine superantigens and attempt to tackle the question of their possible function in bacteria and mice. We present evidence that T-cell stimulation by bacterial superantigens is critical to their toxic properties. In addition, we demonstrate that in mice widespread polymorphisms in superantigens as well as in V_β structural genes results in a state where the mouse population as a whole expresses all V_β elements but that the number of V_β elements expressed in an individual mouse is generally quite limited. We propose that this state may be an evolutionary equilibrium balancing the advantage of a large, diverse T-cell repertoire against the disadvantage of possessing too many potential targets for the microbial superantigen toxins.

RESULTS

Germ-line Limitations on V_β Expression

So far, 22 functional murine V_β elements have been described (Fig. 1). However, no mouse has been found

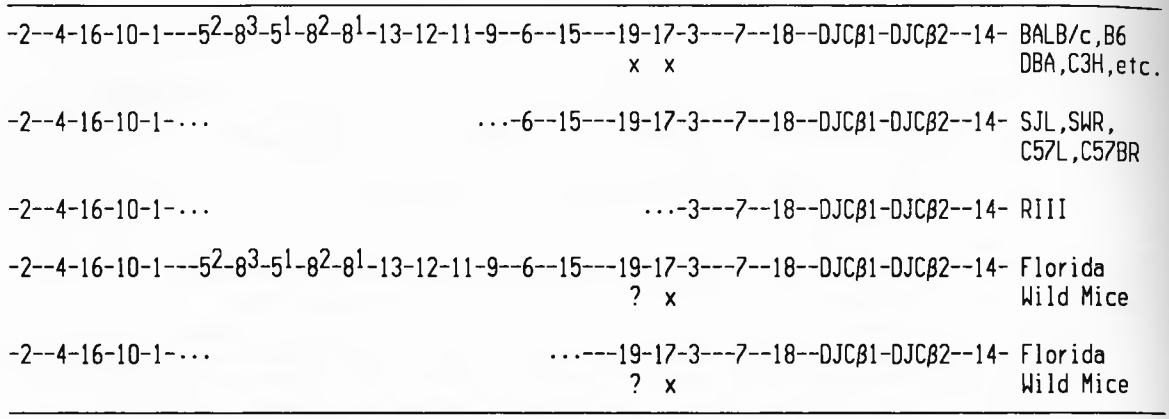

Figure 1. Deletions and mutations within the mouse V_β complex. The genomic organization of the mouse V_β complex is shown for various laboratory strains and for wild mice from central Florida. Approximate position of deletions is shown. (X) V_β elements known to carry point mutations that inactivate the gene. Data are from this paper and Behlke et al. (1986), Malissen et al. (1986), Kappler et al. (1987b), Chou et al. (1987a,b), Lai et al. (1987, 1988), Wade et al. (1988), and Haqqi et al. (1989). Data from V_β19 was from D. Loh (pers. comm.).

that carries all 22 functional V_β genes because various deletions and mutations eliminate at least some V_β elements in all mice examined thus far. Three V_β haplotypes have been identified in laboratory mice. The haplotype carried by most strains has a full complement of V_β genes; however, the V_β17 gene carries a single-base mutation generating a termination codon near the 3′ end of the coding region (Wade et al. 1988), and the V_β19 gene is inactivated by a frameshift caused by a single-base deletion in the leader (D. Loh, pers. comm.). Loh and colleagues have described a large deletion in SJL and a few other strains of mice in the middle of the V_β complex, that eliminates nine functional V_β elements (Chou et al. 1987a). These strains have a functional V_β17 and V_β19 gene. Recently, a similar but more extensive deletion has been found in another laboratory strain, RIII (Haqqi et al. 1989).

Such massive elimination of functional V_β elements should be disadvantageous to mice since the mice carrying these deletions should have a substantially reduced TCR$\alpha\beta$ repertoire. Therefore, one could expect that in the wild the pressure of dealing with environmental pathogens might select against mice carrying the mutations and deletions. On the contrary, Huppi et al. (1988) have found that a deletion similar to that of SJL mice is common in mice from Hebrides. We have examined the V_β complex of a series of 39 wild mice from central Florida. Southern blot analysis of the V_β17 gene in these mice showed a restriction-fragment-length polymorphism pattern identical to the inactive V_β17 allele (Kappler et al. 1987b; Wade et al. 1988), and no T cells were detected in these mice reactive with an antibody specific for V_β17. In addition, a deletion was found in the V_β complex in many of these mice. This deletion was intermediate in length between the two previously described deletions, involving 11 V_β elements (Fig. 1), and was frequent enough in these populations so that 10 of the 39 mice examined were homozygous for the deletion and therefore lacked any T cells bearing the deleted V_β elements (Table 2). Thus, for some reason, mice in the wild tolerate at high-frequency genetic mutations and deletions that limit the TCR$\alpha\beta$ repertoire in some mice.

Deletion of T Cells by Polymorphic V_β-specific Self-superantigens

A similar picture of limited V_β expression has emerged during examination of the V_β-specific self-superantigens. We identified the first antigen of this type as a B-cell product that combined with the MHC class II molecule, IE, to form a ligand for virtually all T cells whose TCR$\alpha\beta$ contained V_β17a (Kappler et al. 1987b; Marrack and Kappler 1988). IE^+ mice carrying the functional V_β17a gene delete virtually all $V_\beta17a^+$ T cells while establishing self-tolerance (Kappler et al. 1987a, 1989a). Since then, we and other investigators have identified the V_β specificity of a number of other self-superantigens. For example, mice carrying the minor lymphocyte-stimulating (Mls-1^a) antigen, long known as a powerful alloantigen, have been shown to eliminate virtually all T cells bearing V_β6 (MacDonald et al. 1988), V_β8.1 (Kappler et al. 1988), and V_β9 (Happ et al. 1989). Likewise, mice carrying the Mls-2^a or Mls-3^a antigen eliminate T cells bearing V_β3 (Pullen et al. 1988, 1989). Examples of these phenomena are shown in Figure 2 and 3, and the properties of the V_β-specific self-superantigens reported so far are summarized in Table 1.

Some of the most striking features of these antigens are the extensive polymorphisms controlling their expression. Thus, in every case, the superantigen and/or the MHC-restricting elements that present it are polymorphic, so that a functional ligand appears in only some strains of mice. For example, the superantigens that delete T cells bearing V_β17a, V_β5, or V_β11 all require an IE molecule for presentation. Therefore, even in the presence of the superantigen, IE^- strains of

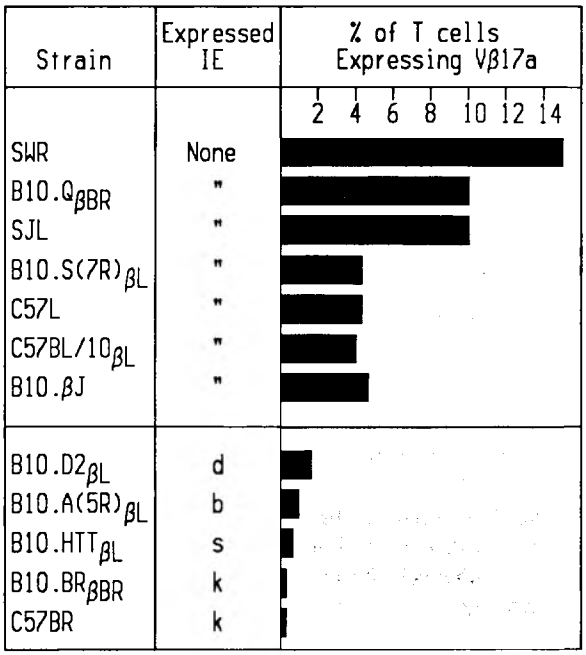

Figure 2. Elimination of V_β17a-bearing T cells in IE$^+$ mice. Peripheral T cells, prepared from a series of mice carrying a functional gene for V_β17, were stained with a monoclonal antibody specific for V_β17 and analyzed by flow cytometry (Kappler et al. 1987b). For each strain, the percentage of T cells bearing V_β17 is shown along with the MHC class II molecules expressed in that strain. Each value shown is an average for those obtained in three or more mice. Standard errors were 10% or less.

mice do not present the antigen, and T cells bearing the relevant V_β elements are not deleted. In the case of Mls-1, Mls-2, and Mls-3, only the *a* allele of these loci encodes a superantigen. The relevant V_β elements are expressed normally in mice carrying the *b* allele. Furthermore, the class II molecule of H-2^q, IA^q, is unable to present these Mls superantigens, so that an H-2^q strain, such as DBA/1 carrying the *a* allele at all three Mls loci, nevertheless expresses normal levels of the relevant V_β elements.

Again, one might suppose that in the wild the antigen and MHC alleles creating superantigen ligands would be selected against because they dramatically limit the TCR$\alpha\beta$ repertoire. To test this idea, we used the available monoclonal antibodies to V_β elements to examine peripheral T cells from the 39 wild mice from Florida mentioned above. We were surprised to find that the pattern of V_β expression in these mice was consistent with the frequent expression of a number of the superantigens listed in Table 1. Figure 4 shows some relevant data from a few of the mice, and the data are summarized in Table 2. In each case, the presence of a functional V_β gene was established by good V_β expression on immature thymocytes (prior to induction of self-tolerance). T cells bearing each of the relevant V_β elements were severely depleted among mature T cells in some of the mice. For example, V_β3-bearing T cells were deleted in most mice, indicating widespread distribution of an Mls-2/3^a-like antigen. Nearly all of these mice expressed an IE molecule, and deletion of V_β5 T cells was very common. The evidence in laboratory mice suggests that the IE-dependent superantigens specific for V_β5 and V_β11 are polymorphic (Bill et al. 1989; Palmer et al., this volume). The results with these wild mice strengthen this conclusion, indicating that different antigens control interactions with these V_β elements and that at least one nonfunctional allele exists for each since the presence of an IE molecule is not sufficient for deletion of T cells bearing these V_β elements and the pattern of deletion for each is different.

The deletion of V_β6- and V_β8.1-bearing T cells in some of these mice indicated the presence of an Mls-1^a-like antigen. Interestingly, the pattern of reactivity with

Table 1. Self-superantigens in the Mouse That Delete T Cells Bearing Particular V_β Elements

Self-superantigen (gene location)	MHC-class-II-presenting elements	V_β elements	References[a]
Nonpolymorphic? B-cell product (unknown locus)	IE	17a	1
Mls-1[a] (chromosome 1)	most IE some IA	6, 8.1, 9	2
Mls-2[a] (single locus)	most IE some IA	3	3
Mls-3[a] (linked to Ly-7)	most IE some IA	3	4
Polymorphic (unknown locus)	IE	5	5
Polymorphic (unknown locus)	IE	11	6
Polymorphic (unknown locus)	most IE some IA?	7	7

[a] (1) Kappler et al. (1987a, b, 1989a); Marrack and Kappler (1988); (2) Kappler et al. (1988); MacDonald et al. (1988); Happ et al. (1989); (3) Pullen et al. (1988, 1989); (4) Palmer et al. (this volume); (5) Bill et al. (1989); and (6) M. Happ et al. (pers. comm.).

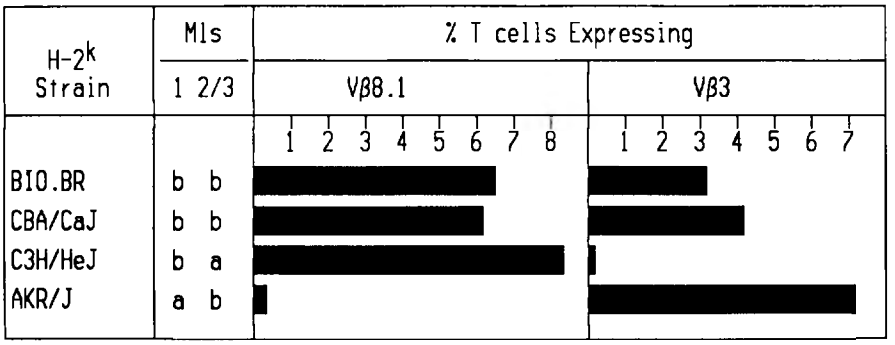

Figure 3. The *a* alleles of the Mls antigens eliminate T cells bearing $V_\beta 8.1$ or $V_\beta 3$. Peripheral T cells from the strains shown are analyzed as in Fig. 2 for expression of $V_\beta 8.1$ or $V_\beta 3$. $V_\beta 8.1$ was analyzed using two monoclonal antibodies, one specific for $V_\beta 8.1$ plus $V_\beta 8.2$ and the other specific for $V_\beta 8.2$ (Kappler et al. 1988). $V_\beta 3$ was analyzed with a specific monoclonal antibody (Pullen et al. 1988).

a series of monoclonal antibodies specific for different combinations of the three members of the $V_\beta 8$ family indicated both that the $V_\beta 8.2$ element in these mice had lost reactivity with one of the antibodies and that T cells bearing this altered $V_\beta 8.2$ were also deleted in Mls-1^a mice (Fig. 4). Consequently, we sequenced this variant $V_\beta 8.2$ and produced a series of T-cell hybridomas from Mls-1^b wild mice bearing the variant $V_\beta 8.2$. The hybridomas indeed showed a very high frequency of reactivity to spleen cells from Mls-1^a-bearing laboratory mice (data not shown), and the sequence showed five amino acid differences from the $V_\beta 8.2$ of laboratory mice, two of which were changes to the corresponding amino acid in the known Mls-1^a reactive $V_\beta 8.1$ gene (Table 3). Therefore, we conclude that the T-cell-deleting alleles of superantigens and the MHC molecules that present them are not selected against in wild mice but rather coexist stably with nondeleting allelic forms. Furthermore, the conversion of the $V_\beta 8.2$ gene to superantigen reactivity suggests that V_β elements may be under selection pressure to be able to recognize superantigens.

Staphylococcus aureus Toxins Are Superantigens in Mice

The data on the distribution of V_β expression in mice suggest that, despite the obvious advantage of a large T-cell repertoire for dealing with a wide variety of pathogens, mice for some reason purposely exist in a state where individuals generally express only a subset of the V_β elements available to the species as a whole. Therefore, one can suppose that there is some offsetting disadvantage to having too many V_β elements expressed in an individual mouse. We have been exploring the possibility that this phenomena is related to

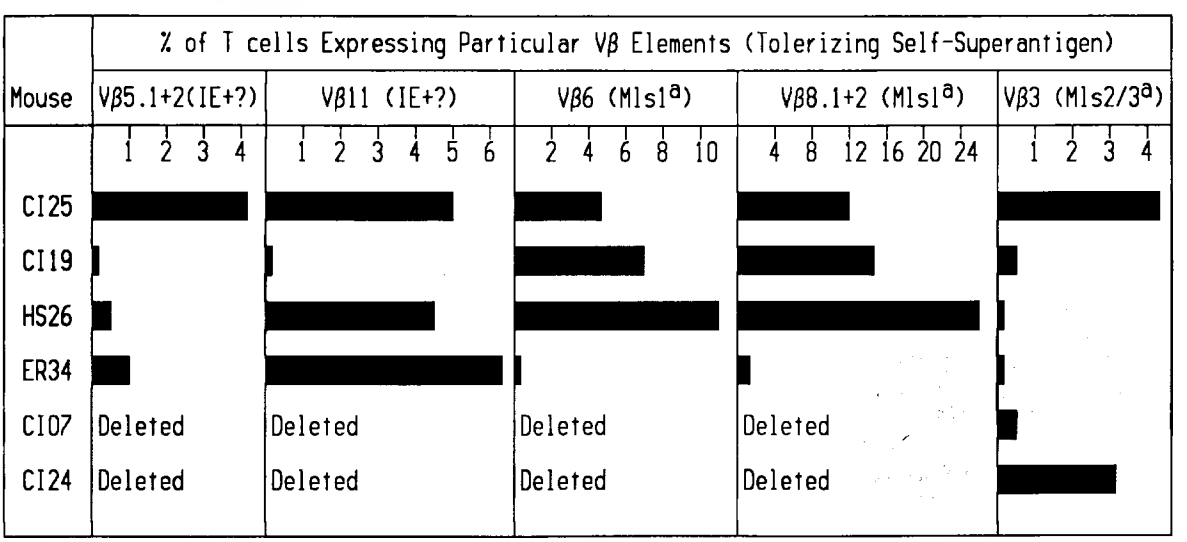

Figure 4. V_β expression in six representative wild mice. Peripheral T cells from six mice taken from three different sites in central Florida were analyzed for V_β expression, as in Fig. 2, using monoclonal antibodies specific for the V_β elements shown. The self-superantigen capable of eliminating T cells bearing these V_β elements are shown in parentheses. In those cases where a value is given, staining of immature cells in the thymus established the presence of a functional V_β structural gene. Those cases marked as "deleted" indicate no staining in the thymus or periphery, indicating the absence of a functional structural gene. In many cases, the deletion of the gene was confirmed by Southern blot analysis.

Table 2. High Frequency of V_β Elimination in Wild Mice

	Mechanism of V_β elimination					
	homozygous gene deletion	homozygous gene mutation	tolerance to self-superantigens			
			Mls-1[a]	Mls-2/3[a]	IE^+?	IE^+?
V_βs eliminated	5.1 + 2 8.1 + 2 + 3 13, 12, 11 9, 6, 15	17	6, 8.1 9 **8.2**	3	5.1 + 2	11
Portion of wild mice with this elimination	10/39	39/39	8/29	32/39	21/29	2/29

toxic microbial products that are themselves V_β-specific superantigens and use this property in their mode of action.

A number of microbial products have been identified as potent T-cell stimulators in vitro in mouse and man. The related exotoxins of *S. aureus* are particularly good examples (Bergdoll 1983; Couch et al. 1988). In humans, these toxins are responsible for a large portion of food poisoning cases (staphylococcal enterotoxins A to E [SEA to SEE]), as well as other diseases such as scalded skin syndrome (exfoliating toxin) and toxic shock syndrome (toxic shock syndrome toxin [TSST-1]). Originally considered nonspecific mitogens, recent work has shown that these toxins must bind MHC molecules in order to stimulate T cells (Fischer et al. 1989; Fraser 1989) and that for each toxin only T cells bearing particular V_β elements respond (Choi et al. 1989; Janeway et al. 1989; Kappler et al. 1989b; White et al. 1989). Table 4 summarizes our results in mice using some of the *S. aureus* toxins. We found that each toxin stimulates a different set of V_β-bearing T cells. Even very closely related toxins (Couch et al. 1988), such as SEB and SEC1 or SEA and SEE, stimulate different T cells. Since most of the toxins stimulate T cells bearing any of several V_β elements, the proportion of total T cells stimulated is in some cases very high. For example, in the mouse SEB stimulates T cells bearing V_β3, V_β8, V_β7, or V_β17a. Depending on the strain of mouse, this can account for over 30% of all T cells. Similarly, in humans, we have found that these toxins stimulate T cells bearing particular V_β elements and that particular toxins often stimulate T cells bearing homologous V_β elements in mice and humans (Choi et al. 1989).

T Cells Bearing Specific V_βs Required in the Mode of Action of SEB

Despite the considerable variability of the primary sequences of these toxins, their ability to act as superantigens appears to be a conserved property. Furthermore, since each has a different V_β specificity, their superantigen function appears not to be an accidental consequence of the conservation of some common structural element required for some other function. Therefore, it is reasonable to suppose that the pathology associated with these toxins (e.g., vomiting, diarrhea, skin rashes, and shock) may be an indirect consequence of the massive in vivo activation of T cells and release of powerful lymphokine mediators rather than a direct effect of the toxins on the target tissue. This idea is difficult to test directly in humans where most of the pathology associated with these toxins has been identified so we have begun to study the effects of these toxins in mice in vivo.

We have begun our experiments with SEB. Mice given a single injection of SEB develop a shock-like syndrome within 24 hours. Usually, high doses are required ($> 50\ \mu g$/mouse), correlating with the observation that this toxin, which was isolated from a strain of Staphylococcus that infects humans, must be used at a higher concentration in vitro to stimulate murine T cells than human T cells. The most obvious in vivo symptom in mice from this toxin is extremely rapid weight loss with wasting and in some cases death within 2–3 days. Those mice that survive for 3 days usually recover completely over the next few days. We have used this simple assay to assess the importance of the superantigen properties of SEB for its in vivo toxicity.

Table 3. Amino Acid Substitutions in the V_β8.2 Gene of Wild Mice

Mouse	V_β	Mls-1[a] reactive	Binds MAb F23.2	Amino Acid				
				8	22	51	70	71
C57BL/6	8.2	–	+	N	N	G	E	N
Wild	8.2	+	–	**S**	D	**D**	K	E
C57BL/6	8.1	+	–	**S**	**H**	**D**	E	N

C57BL/6 sequences from Chou et al. (1987b). Amino acid numbering starts at the alanine at the amino termi of the mature proteins.

Table 4. *S. aureus* Toxins Are Superantigens in the Mouse

S. aureus toxin	Response of T cells bearing V_β																	
					5				8									
	1	2	3	4	1	2	6	7	1	2	3	10	11	12	14	15	16	17·
SEA	+	−	+	−	−	−	−	−	−	−	−	−	+	−	−	−	−	+
SEE	−	−	−	−	−	−	−	−	−	−	−	−	+	−	−	+	−	n.d.
SEB	−	−	+	−	−	−	−	+	+	+	+	−	−	−	−	−	−	+
SEC1	−	−	+	−	−	−	−	−	−	+	−	−	−	−	−	−	−	+
TSST	−	−	+	−	−	−	−	−	−	−	−	−	−	−	−	+	−	+

V_β usage was analyzed in T-cell blasts after stimulation with each of the toxins shown. Most data are from B10.BR mice (H-2^k). To analyze V_β elements that recognize IE-dependent superantigens IE^- mice were used: $B10.Q_{\beta BR}$ mice for $V_\beta 17$ and B10.Q mice for $V_\beta 5$ and $V_\beta 11$. Staining with V_β-specific monoclonal antibodies was used where possible, otherwise RNA was prepared from each of the blast populations and V_β expression established using a quantitative dot blot. Either unstimulated (staining) or concanavalin-A-stimulated T-cell blasts (RNA dot blots) were used as control cells. Pluses indicate an enrichment in V_β expression in the toxin-stimulated T-cell blasts compared with the control cells of twofold or better. n.d. indicates not determined.

Nude mice and neonatal mice, both of which lack mature functional T cells, are extremely resistant to SEB (data not shown), implicating T cells in the mode of action of SEB. However, the hypothesis we wished to test was that the various mechanisms that mice use to limit V_β expression may be a protective mechanism to limit the possible targets for these toxins in vivo. Therefore, we devised an experiment to test this idea more directly. We bred mice that combined a genetic deletion with the appropriate tolerizing V_β-specific self-superantigens to lower drastically the levels of the T cells bearing the V_β elements recognizing SEB. One strain, B10.BR(βBR) contributed a genetic deletion involving all members of the $V_\beta 8$ family. The other (CBA/J) contributed the self-superantigens deleting $V_\beta 3^+$ (Mls-2^a or Mls-3^a) and $V_\beta 7^+$ (unknown locus) T cells. (B10.BR[βBR] × CBA/J)F_1 mice were bred with B10.BR(βBR) mice and the progeny screened for the homozygous gene deletion and heterozygous but dominant superantigens. These mice occurred approximately with the predicted 1 in 8 frequency. On average, the sum of $V_\beta(3 + 7 + 8)$ T cells in these mice was less than 6% compared with 32% in control B10.BR mice, which lack both the genetic deletion and the tolerizing self-superantigens. All mice in these experiments lacked $V_\beta 17^+$ T cells both because they expressed the IE-molecule of H-2^k and in some cases because they bore the inactive $V_\beta 17$ gene. The experimental and control mice were injected with SEB, and their weight was followed over the next several days (Table 5). Whereas the control mice with a high level of SEB-reactive T cells showed a typical response with rapid weight loss, the mice specifically low in SEB-reactive T cells were protected against the toxin.

These results demonstrate that V_β-specific T-cell stimulation is important in the mode of action of these toxins in mice. The advantage to the bacteria of massive T-cell activation is not immediately apparent; however, our preliminary experiments have shown that after SEB administration mice temporarily produce a poor T-cell response to other antigens, which may be of some advantage to the bacteria producing these toxins.

IMPLICATIONS

Our experiments establish that in mice the selective advantage of a large, diverse TCR$\alpha\beta$ repertoire is not strong enough to weed out any of the many polymorphisms that limit V_β expression in laboratory and wild mice. Rather, they suggest an equilibrium in which mice exist in a state where all V_β elements are available to the species as a whole, but individuals express only a subset of the total. This implies some selective disadvantage to the expression of too many V_β elements in one individual. We suggest that this disadvantage lies in the susceptibility of mice to microbial toxins that rely on the massive stimulation of T cells bearing particular V_β elements for their toxicity. Our finding that mice constructed to have the right combination of V_β gene deletions and tolerizing V_β-specific self-superantigens are resistant to the appropriate toxin is consistent with this view.

Our results imply that the V_β repertoire of the mouse population is very flexible, capable of changing rapidly to accommodate the threat either from a new pathogen, the immune response to which may require a particular V_β element, or from a new microbial toxin in which case elimination of particular V_β elements may

Table 5. Deletion of T Cells Bearing Approximate V_β Elements Protects Mice from SEB

Mouse	T cells bearing $V_\beta 3$, 8, or 7 (%)	Weight loss following SEB (%) day 1	day 2	day 3
B10.BR	32	10.9 ±0.7	9.7 ±1.0	5.3 ±1.6
(B10.Q[βBR] × CBA/J)F_1 × B10.Q(βBR)	**6**	2.6 ±1.4	−0.2 ±1.1	2.6 ±1.1

Mice were given 50 μg of SEB intraperitoneally on day 0.

be protective. The self-superantigens are a particularly efficient way of limiting expression of particular V_β elements although keeping them in reserve for rapid reexpression should the need arise. Suppression of a particular V_β is achieved without altering the V_β structural gene in mice heterozygous for the relevant superantigen. Mice expressing V_β appear in every mating between superantigen heterozygous mice. For example, to maintain the lack of $V_\beta 6$ expression in 90% of mice, a V_β haplotype with a $V_\beta 6$ gene deletion must occur at a frequency of 95% in the gene pool, and the reemergence of $V_\beta 6$ can only come from matings involving the 10% of mice already expressing $V_\beta 6$. On the other hand, lack of $V_\beta 6$ expression can be maintained at the 90% level with a frequency of only 68% of the Mls-1^a gene, and approximately 30% of matings, including those between two Mls-1^a heterozygous mice each of which lacks $V_\beta 6$ expression, will yield at least some $V_\beta 6$-expressing progeny.

ACKNOWLEDGMENTS

We thank Mike Bevan, Uwe Staerz, Jerry Bill, Osami Kanagawa, Ed Palmer, and Craig Okada for providing anti-V_β monoclonal antibodies. We thank also Ella Kushnir, Terri Wade, and Rhonda Richards for their excellent technical assistance. This work was supported in part by U.S. Public Health Service grants AI-17134, AI-18785, and AI-22295.

REFERENCES

Behlke, M., H. Chou, K. Huppi, and D. Loh. 1986. Murine T cell receptor mutants with deletions of β-chain variable region genes. *Proc. Natl. Acad. Sci.* **83:** 767.

Bergdoll, M. 1983. Enterotoxins. In *Staphylococci and staphylococcal infections* (eds. C.S.F. Easman and C. Adlam), p. 559. Academic Press, New York.

Bevan, M. and P. Fink. 1978. The influence of thymus H-2 antigens on the specificity of maturing killer and helper cells. *Immunol. Rev.* **42:** 3.

Bill, J., V. Appel, and E. Palmer. 1989. The MHC molecule I-E is necessary, but not sufficient for the clonal deletion of Vβ11 bearing T cells. *J. Exp. Med.* **169:** 1405.

Blackman, M.A., P. Marrack, and J. Kappler. 1989. Influence of the MHC on positive thymic selection of $V_\beta 17a^+$ T cells. *Science* **244:** 214.

Choi, Y., B. Kotzin, L. Herron, J. Callahan, P. Marrack, and J. Kappler. 1989. Interaction of S. aureus toxin superantigens with human T cells. *Proc. Natl. Acad. Sci.* **86:** 8941.

Chou, H., C. Nelson, S. Godambe, D. Chaplin, and D. Loh. 1987a. Germline organization of the murine T cell receptor β-chain genes. *Science* **238:** 545.

Chou, H., S. Anderson, M. Louie, S. Godambe, M. Pozzi, M. Behlke, K. Huppi, and D. Loh. 1987b. Tandem linkage and unusual RNA splicing of the T cell receptor β-chain variable-region genes. *Proc. Natl. Acad. Sci.* **84:** 1992.

Couch, J., M. Soltis, and M. Betley. 1988. Cloning and nucleotide sequence of the type E staphylococcal enterotoxin gene. *J. Bacteriol.* **170:** 2954.

Fischer, H., M. Dohlsten, M. Lindvall, H. Sjögren, and R. Carlsson. 1989. Binding of staphylococcal enterotoxin A to HLA-DR on B cell lines. *J. Immunol.* **142:** 3151.

Fraser, J.D. 1989. High-affinity binding of staphylococcal enterotoxins A and B to HLA-DR. *Nature* **339:** 221.

Happ, M.P., D. Woodland, and E. Palmer. 1989. A third T cell receptor V_β gene encodes reactivity to Mls-1^a gene products. *Proc. Natl. Acad. Sci.* (in press).

Haqqi, T., S. Banerjee, G. Anderson, and C. David. 1989. RIII S/J (H-2^r). An inbred mouse strain with a massive deletion on T cell receptor V_β genes. *J. Exp. Med.* **169:** 1903.

Huppi, K., B. D'Hoostelaere, B. Mock, E. Jouvin-Marche, M. Behlke, H. Chou, R. Berry, and D. Loh. 1988. T-cell receptor VTβ genes in natural populations of mice. *Immunogenetics* **27:** 51.

Janeway, C.A., Jr., J. Yagi, P. Conrad, M. Katz, S. Vroegop, and S. Buxser. 1989. T cell responses to Mls and to bacterial proteins that mimic its behavior. *Immunol. Rev.* **107:** 61.

Kappler, J.W., E. Kushnir, and P. Marrack. 1989a. Analysis of Vβ17a expression in new mouse strains bearing the Vβ^a haplotype. *J. Exp. Med.* **169:** 1533.

Kappler, J., N. Roehm, and P. Marrack. 1987a. T cell tolerance by clonal elimination in the thymus. *Cell* **49:** 273.

Kappler, J.W., U. Staerz, J. White, and P.C. Marrack. 1988. Self-tolerance eliminates T cells specific for Mls-modified products of the major histocompatibility complex. *Nature* **332:** 35.

Kappler, J., T. Wade, J. White, E. Kushnir, M. Blackman, J. Bill, R. Roehm, and P. Marrack. 1987b. A T cell receptor Vβ segment that imparts reactivity to a class II major histocompatibility complex product. *Cell* **49:** 263.

Kappler, J., B. Kotzin, L. Herron, E. Gelfand, R. Bigler, A. Boylston, S. Carrel, D. Posnett, Y. Choi, and P. Marrack. 1989b. V_β-specific stimulation of human T cells by staphylococcal toxins. *Science* **244:** 811.

Kisielow, P., H. Teh, H. Blüthmann, and H. von Boehmer. 1988. Positive selection of antigen-specific T cells in thymus by restricting MHC molecules. *Nature* **335:** 730.

Lai, E., R. Barth, and L. Hood. 1987. Genomic organization of the mouse T-cell receptor β-chain gene family. *Proc. Natl. Acad. Sci.* **84:** 3846.

Lai, E., P. Concannon, and L. Hood. 1988. Conserved organization of the human and murine T cell receptor β-gene families. *Nature* **331:** 543.

MacDonald, H.R., R. Schneider, R.K. Lees, R.C. Howe, H. Acha-Orbea, H. Festenstein, R.M. Zinkernagel, and H. Hengartner. 1988. T-cell receptor V_β use predicts reactivity and tolerance to Mls^a-encoded antigens. *Nature* **332:** 40.

Malissen, M., C. McCoy, D. Blanc, J. Trucy, C. Devaux, A. Schmitt-Verhulst, F. Fitch, L. Hood, and B. Malissen. 1986. Direct evidence for chromosomal inversion during T-cell receptor β-gene rearrangements. *Nature* **319:** 28.

Marrack, P. and J. Kappler. 1988. T cells can distinguish between allogeneic major histocompatibility complex products on different cell types. *Nature* **332:** 6167.

Pullen, A.M., P. Marrack, and J.W. Kappler. 1988. The T cell repertoire is heavily influenced by tolerance to polymorphic self antigens. *Nature* **335:** 796.

———. 1989. Evidence that Mls-2 antigens which delete $V_\beta 3^+$ T cells are controlled by multiple genes. *J. Immunol.* **142:** 3033.

Sha, W., C. Nelson, R. Newberry, D. Kranz, J. Russell, and D. Loh. 1988. Positive and negative selection of an antigen receptor on T cells in transgenic mice. *Nature* **336:** 73.

Wade, T., J. Bill, P.C. Marrack, E. Palmer, and J.W. Kappler. 1988. Molecular basis for the nonexpression of Vβ17 in some strains of mice. *J. Immunol.* **141:** 2165.

White, J., A. Herman, A.M. Pullen, R. Kubo, J. Kappler, and P. Marrack. 1989. The Vβ-specific superantigen staphylococcal enterotoxin B: Stimulation of mature T cells and clonal deletion in neonatal mice. *Cell* **56:** 27.

Zinkernagel, R., G. Callahan, A. Althage, S. Cooper, P. Klein, and J. Klein. 1978. On the thymus in the differentiation of H-2 self-recognition by T cells: Evidence for dual recognition? *J. Exp. Med.* **147:** 882.