A murine macrophage line of the H-2^{d/f} haplotype can activate H-2^k suppressor T cells

(antigen-presenting cell/suppression/glycosylation)

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ABSTRACT Little is known about the molecular basis for activation of suppressor T cells. In this report we describe two macrophage cell lines, BAC1.2.SC8 and its variant progeny B26, that differ in their ability to activate suppressor T cells. The SC8 line is derived from a (BALB/c × A.CA)F₁ (H-2^{d/f}) mouse and is haploid with respect to I-E^d. It is capable of activating I-E^d-restricted helper T cells as well as poly-(Glu⁵⁰Tyr⁵⁰)-specific I-E^d-restricted suppressor cells. The B26 variant can activate H-2^d-restricted helper T cells but activates H-2^k-restricted suppressor cells. The I-E^d molecules of SC8 and of B26 have identical amino acid sequences. This suggests that suppressor T cells either recognize posttranslational modifications of the I-E molecule or that there is another accessory molecule that helps determine the major histocompatibility complex restriction in the activation of suppressor T cells.

The past decade has seen a large number of studies aimed at understanding the molecular basis for immunologic nonresponsiveness. These studies have shown that in some instances nonresponsiveness is the consequence of a lack of presentation of antigen by antigen-presenting cells or a lack of helper T cells specific for the antigen (1–3). In some instances, however, the lack of an immune response is mediated by the presence of regulatory or suppressor T cells (4–6).

A variety of data suggest that, like helper T cells, suppressor T cells require an antigen-presenting cell for activation and that the interaction of suppressor T cells with the antigen-presenting cell is genetically restricted (7, 8). Antigen-presenting cells and suppressor T cells must be matched at the class II locus of the major histocompatibility complex (MHC). Additionally, antibodies to one class II gene product, I-E, have been shown to block activation of suppressor cells, suggesting that class II gene products play a direct role in the activation of suppressor cells (9). Suppressor T cells possess a membrane determinant I-J; by using congenic inbred strains of mice, the I-J determinant was initially mapped to the class II gene locus within the MHC (10). Molecular genetic studies of the class II gene locus, however, showed that no I-J genes are present there (11). An alternative explanation for the mapping of I-J to the MHC is that I-J determinants are selected by MHC class II antigens (12, 13). Although I-J determinants could then be encoded anywhere in the genome, only those T cells expressing I-J determinants that can interact with the class II molecules on antigen-presenting cells would mature in the thymus and be present as immunocompetent T cells in peripheral lymphoid organs. The class II molecules would, therefore, select the expressed I-J determinant. Particular I-J molecules would be expressed in mice with particular class II molecules, and I-J would appear to map to the MHC. To explore this hypothesis, we generated

anti-idiotypes to anti-I-J antibodies and showed that these reagents bound to antigen-presenting cells and inhibited activation of suppressor T cells in a genetically restricted fashion (14, 15). We hypothesized the existence of an I-J interacting molecule that might be encoded within the MHC. Concurrent experiments in chimeric and transgenic mice showed that a mouse can acquire a new I-J phenotype when given a new I-E molecule (12, 16). These experiments suggested that the I-J phenotype is acquired by a process of selection during the ontogeny of the immune system and that I-E is at least part of the selecting principle.

While the experiments described above led to the suggestion that the I-E molecule plays an important role in the activation of suppressor T cells, the I-E molecule alone cannot be solely responsible for mediating the nonresponsiveness of peripheral blood T cells. The inbred strains of mice B10.A(5R) and B10.A(3R) both express the same I-E molecule, I-E^k. The sequences of the β_1 exon of the E β chain in these two strains are presumed to be identical, and the sequences of the β_2 exons in these two strains are identical, so the amino acid sequence of the I-E molecules must also be the same (17). Furthermore, no polymorphisms of the E α chain genes in these two strains have been found (18). Yet, T cells from B10.A(5R) and B10.A(3R) mice express different I-J phenotypes (10). The I-J phenotype, therefore, must reflect more than the amino acid sequence of the I-E molecule. The B10.A(5R) and B10.A(3R) strains must differ either in posttranslational modifications of the I-E molecule or in a second, as yet unidentified, molecule that mediates interactions between antigen-presenting cells and suppressor T cells.

Here, we report a mutant macrophage cell line that has an I-E^d molecule with an identical amino acid sequence to the parental line but behaves differently with respect to activation of suppressor T cells specific for a random copolymer consisting of 50% glutamic acid and 50% tyrosine [poly(Glu⁵⁰-Tyr⁵⁰)]. The parental line BAC1.2.SC8 (H-2^{d/f}) can present poly(Glu⁵⁰Tyr⁵⁰)-specific T-suppressor factor 1 [poly(Glu⁵⁰-Tyr⁵⁰)-TsF1], as expected, to cells of the d haplotype, whereas the variant, B26, has lost this ability and instead can activate poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor T cells of the k haplotype.

MATERIALS AND METHODS

Mice. BALB/c and BALB/k mice were a gift from Frank Lilly (Albert Einstein College of Medicine, Bronx, NY). B10.A(5R), B10.A(3R), and AKR mice were obtained from The Jackson Laboratory.

Reagents. Ovalbumin, ethyl methanesulfonate, and cytochrome c were purchased from Sigma. poly(Glu⁵⁰Tyr⁵⁰)

(Miles, Yeda Rehovot, Israel) was coupled to methylated bovine serum albumin as previously described (9).

Cell Lines. BAC1.2 is a cloned murine macrophage line generated by simian virus 40 transfection of spleen cells from a (BALB/c $\times$ A.CA)F₁ (H-2^{d/f}) mouse and is able to present antigen to T cells in an MHC-restricted manner (19). SC8 is a subclone of BAC1.2 with high level expression of Ia molecules by fluorescence analysis. The SC8 cells are grown in Dulbecco's modified Eagle's medium (DMEM) with 15% (vol/vol) fetal calf serum (HyClone), 1% penicillin and streptomycin, 1% nonessential amino acids (GIBCO), 10% NCTC 109 (M.A. Bioproducts), and 20% (vol/vol) L-cell supernatant as source of colony-stimulating factor 1. The T-cell hybridomas 3D026.1 (I-E^d and ovalbumin specific), 3D062.1 (I-E^d specific), and D011.10 (I-A^d and ovalbumin specific) were gifts from J. Kappler and P. Marrack (National Jewish Hospital, Denver). The 2H10 (I-E^k and cytochrome *c* restricted) T-cell line was obtained from R. Schwartz (National Institutes of Health, Bethesda, MD) (20). Macrophage hybridoma line 63 (H-2^{d/k}) was obtained from M. Dorf (Harvard Medical School, Boston). The T-cell hybridomas and the 63 cell line were grown in DMEM with 10% fetal calf serum, 1% glutamine, and 1% penicillin and streptomycin.

The interleukin 2-dependent cytotoxic T-cell line CTLL-1 was obtained from G. Ju (Hoffmann-LaRoche, Nutley, NJ) and is grown in RPMI 1640 (GIBCO) with 10% fetal calf serum, 1% penicillin and streptomycin, 1% glutamine, 20 mM Hepes (Sigma), 50 μ M 2-mercaptoethanol, and 20% concanavalin A-induced mouse spleen cell supernatant.

Antigen-Presenting Assay. T hybridoma cells were incubated with various numbers of macrophages and the appropriate antigen at 0.2 mg/ml in 200 μ l for 24 hr. The supernatants were then transferred to another 96-well plate containing CTLL-1 cells and assayed for interleukin 2 content by determining the incorporation of [³H]thymidine of the interleukin 2-dependent CTLL-1 cells.

Hemolytic Plaque Assay for Activation of Suppressor T Cells. The poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor T-cell line WF11.3A1 was a gift from C. Waltenbaugh (Northwestern University Medical School, Chicago) (21). It secretes an I-J-bearing monoclonal poly(Glu⁵⁰Tyr⁵⁰)-TsF1 and can activate a suppressor T-cell cascade in both the H-2^d and H-2^k haplotype.

Nonconfluent cultures of macrophages in 24-well plates (Linbro) were washed three times with chilled DMEM. Then 1 ml of culture supernatant containing poly(Glu⁵⁰Tyr⁵⁰)-TsF1 was added to the adherent macrophages, and they were incubated for 15 min on ice. The cells were washed, detached from the tissue culture plates with a rubber policeman, counted, and added to the spleen cell culture. A single-cell suspension of spleen cells was placed in a modified Mishell-Dutton type culture. Fifty microliters of the antigen-pulsed or nonpulsed macrophage cell suspension (8000 cells) and 2.5 μ g of poly(Glu⁵⁰Tyr⁵⁰) as poly(Glu⁵⁰Tyr⁵⁰) coupled to methylated bovine serum albumin were added per well. As a positive control for suppression, a 1:100 dilution of poly(Glu⁵⁰Tyr⁵⁰)-TsF1 was added. In some cultures 1 ml of antibodies at 10 μ g/ml was added. All the culture conditions as well as the plaque-forming cell assay have been described (21).

Construction of a Genomic Library and Sequence Determination of the I-E^d Molecule. Total DNA was extracted from B26 cells and partially digested with *Mbo* I at an enzyme-to-substrate ratio of 0.3:1 for 20 min at 37°C. DNA from 23 to 6 kilobases was fractionated on a sucrose gradient and ligated to the EMBL3 vector at the *Bam*HI site. The phage clones were screened with a ³²P-labeled *Pst* I fragment containing E β cDNA or an *Eco*RI fragment containing E α cDNA (gifts from R. Germain, National Institutes of Health). Phage DNA from the positive clones was analyzed by Southern blots, and the clones containing the whole E β or E α gene

were used for subcloning. All coding exons of the E α ^d and E β ^d were subcloned into either the pGEM 2 vector or the M13 phage vector and sequenced by the dideoxy chain termination method with T7 DNA polymerase (Sequenase, United States Biochemical).

cDNA Sequence of the I-E^d Molecule of B26. Total RNA from frozen B26 cells was extracted by using guanidinium isothiocyanate (22). Complementary DNA was synthesized from 5 μ g of total RNA with 21-base-pair oligomers that specifically bind to the 3' untranslated region of E β ^d or E α ^d gene. The cDNAs were then used as templates to synthesize double-stranded DNA by polymerase chain reaction with another 21-base-pair oligomer that specifically binds to the 5' leader region of each mRNA. These double-stranded DNAs were cloned into the pGEM 2 vector at the *Sma* I site and sequenced by the dideoxy chain termination method.

Generation of Anti-B26 Antibodies. BALB/c mice, at 4–6 weeks of age, were immunized i.p. with 1×10^6 SC8 cells. Twenty-four and 48 hr later, they received an i.p. injection of cyclophosphamide (Sigma; 2 μ g/ml of saline) at 100 μ g/kg of body weight. This treatment was repeated every 2 weeks for 6 consecutive weeks (23). The mice were allowed to rest for 6 weeks before they were immunized i.p. with 1×10^6 B26 cells and boosted 2 weeks later. Three days after the last immunization, the mice were sacrificed and the spleens were fused with X63 Ag 8.653 cells. Wells producing anti-B26 specific antibodies were identified with an ELISA technique, and cells from these wells were cloned.

RESULTS

We have previously reported the characterization of a murine macrophage line, BAC1.2, derived from splenic monocytes of a [BALB/c (H-2^d) $\times$ AC.A (H-2^f)]F₁ mouse. The AC.A mouse is of the H-2^f haplotype. Because it expresses neither the α nor the β chain of an MHC class II I-E molecule, there is no complementation in the F₁ animal. The BAC1.2 cell line therefore is haploid with respect to I-E and has only I-E^d on its surface. SC8 is a subclone of BAC1.2 that has characteristic macrophage properties with high-level expression of Ia molecules and can present antigen to helper T cells in an MHC-restricted manner. We have previously shown that the BAC1.2 line can activate poly(Glu⁵⁰Tyr⁵⁰)-specific suppression and present poly(Glu⁵⁰Tyr⁵⁰)-TsF1 to primed BALB/c spleen cells (15). Anti-I-E antibodies block BAC1.2-mediated induction of suppression. SC8 can also serve as an accessory cell in generating poly(Glu⁵⁰Tyr⁵⁰)-specific suppression in the d haplotype, and anti-I-E^d antibodies can block this activation (data not shown). These data indicate that I-E^d on the membrane of SC8 interacts with suppressor T cells in the poly(Glu⁵⁰Tyr⁵⁰) system.

SC8 cells were then treated with ethyl methanesulfonate to generate mutants. Because SC8 is functionally haploid at the I-E locus, mutations of the I-E molecule can be easily detected. Our initial intention was to make macrophage variants that can no longer present antigen to helper T cells. The selection strategy used has been described (19). All surviving clones were examined for their ability to present poly(Glu⁵⁰Tyr⁵⁰)-TsF1 to d haplotype-restricted T suppressor cells as well as a variety of antigens to appropriate I-E^d-restricted helper T lines. One variant, B26, has lost the ability to activate poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor cells of the H-2^d haplotype (Table 1). It has, however, no identified defect in presenting antigen to I-E^d-restricted helper T cells (Table 2). In an effort to determine if this might reflect an altered density of I-E^d on the cell surface, we examined the membrane expression of I-E^d on B26 by using indirect immunofluorescence with anti-I-E^d antibodies and found that there was no difference in I-E^d expression between B26 and SC8 (data not shown).

Table 1. The B26 (H-2^{d/f}) variant activates suppressor T cells of the H-2^k haplotype

Cells	Poly(Glu ⁵⁰ Tyr ⁵⁰)-TsF1	Poly(Glu ⁵⁰ Tyr ⁵⁰)-specific plaque-forming cells per culture		
		BALB/c (H-2 ^d)	AKR (H-2 ^k)	BALB/k (H-2 ^k)
SC8	—	3040 ± 98	2700 ± 116	2180 ± 420
	+	0 ± 100	3420 ± 473	2640 ± 820
B26	—	1880 ± 140	3440 ± 150	2980 ± 320
	+	2520 ± 430	0 ± 889	0 ± 180

All the cultures contained 5 µg of poly(Glu⁵⁰Tyr⁵⁰) coupled to methylated bovine serum albumin. One representative experiment of three is shown, and the results are expressed as the mean ± SE. Values in boldface italic type show suppression.

Since B26 lost its ability to present poly(Glu⁵⁰Tyr⁵⁰)-TsF1 to H-2^d suppressor cells, we wanted to examine whether B26 could present poly(Glu⁵⁰Tyr⁵⁰)-TsF1 to suppressor cells of another haplotype. To our surprise, B26 has acquired the ability to activate poly(Glu⁵⁰Tyr⁵⁰)-specific suppression in AKR (H-2^k) and BALB/k (H-2^k) spleen cells; the parental line SC8 can present poly(Glu⁵⁰Tyr⁵⁰)-TsF1 only to BALB/c spleen cells (Table 1). As both B10.A(5R) and B10.A(3R) mice express I-E^k, but have different suppressor T-cell phenotypes, we examined whether B26 could act as an accessory cell for B10.A(5R) and B10.A(3R) suppressor T cells. The results indicate that B26 can activate poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor T cells from the B10.A(5R) strain (I-J^k) but not the B10.A(3R) strain (I-J^b), whereas the parental SC8 line does not activate suppression in either strain (Table 3). This property of B26 cells represents a stable phenotype that is not altered with interferon γ treatment (data not shown).

To determine if k haplotype specificities had been more generally acquired, we assayed B26 for its ability to activate an I-E^k-restricted helper T hybridoma line and found that B26 cannot present antigen to the cytochrome c-specific I-E^k-restricted T-cell line 2H10 (Table 4).

We believe that a possible interpretation of the data is that B26 possessed a I-E molecule that was altered in such a way that it can no longer be recognized by H-2^d-specific suppressor T cells but can be recognized by H-2^k-specific suppressor T cells. We therefore cloned and sequenced the E_α^d and E_β^d genes from a genomic library of B26. Two 16-kilobase fragments containing the E_α^d and E_β^d genes were isolated and subcloned into plasmid or phage vectors. All coding exons were sequenced from both directions by using oligomers specific for the flanking regions at the 3' and 5' end of each exon. Both the E_α^d and E_β^d genes proved to be unmutated. To ensure that there was not a splicing mutation giving rise to a variant mRNA, the polymerase chain reaction was used on cell line RNA to obtain E_α^d and E_β^d cDNA fragments. The junctional regions were sequenced. The cDNA was likewise wild type. By extrapolation, the I-E^d molecule of B26, therefore, must have a normal amino acid sequence.

To determine the nature of the molecule responsible for the change in haplotype specificity in the poly(Glu⁵⁰Tyr⁵⁰) suppression system, we generated a panel of antibodies with differential recognition of B26 and its parental line, SC8. Mice immunized with SC8 cells were treated with cyclophosphamide to kill all activated anti-SC8 lymphocytes. The mice were then immunized with B26 cells (details in *Materials and Methods*). This procedure allows activation of only those B cells that recognize epitopes uniquely expressed on B26. Hybridomas were then made from spleen cells of the immunized mice and screened with an ELISA technique. All antibodies with specific recognition of B26 cells were tested for their ability to block B26-mediated activation of H-2^k suppressor T cells. Two anti-B26 antibodies, 2F6 and 2E7, can specifically inhibit B26 activation of poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor T cells in the k haplotype but not SC8 activation of H-2^d suppressor cells. These antibodies can also block the suppression induced by H-2^k splenic accessory cells (Table 5). The ability of B26 to activate I-E^d-restricted helper cells is not blocked by these two antibodies, but it is blocked by a conventional anti-I-E antibody (data not shown).

DISCUSSION

We have generated a H-2^{d/f} macrophage cell line, SC8, which can present poly(Glu⁵⁰Tyr⁵⁰)-TsF1 to suppressor T cells in a genetically restricted fashion. The I-E molecule on SC8 is essential for this function because activation of poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor T cells can be blocked by anti-I-E^d antibodies.

B26, an *in vitro* variant of SC8, has the ability to present poly(Glu⁵⁰Tyr⁵⁰)-TsF1 to suppressor T cells of the k haplotype and not of the d haplotype. This cell line thus offers an opportunity to study the genetic restriction of activation of suppressor T cells in an antigen-specific system. Since it is clear that poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor T cells interact with the I-E molecule on antigen-presenting cells, we chose to examine the structure of the I-E molecule of B26 cells. We first sequenced the coding exons of the I-E α- and β-chain genes. There is no mutation in these genes. Since the human class II molecules, HLA-DR, was found to have two different membrane forms resulting from different mRNA splicing (24), we sequenced the B26 I-E molecule at the cDNA level. No abnormal splicing was found. Therefore, the I-E molecule of B26 has an amino acid sequence identical to the I-E molecule of the parental SC8 cells. This is an *in vitro* analogue of the B10.A(5R)/(3R) paradigm showing that accessory cells with an identical I-E molecule can activate suppressor T cells of different haplotypes. Because B26 activates poly(Glu⁵⁰Tyr⁵⁰)-specific suppressor T cells from the B10.A(5R) mouse (I-J^k) and not the B10.A(3R) mouse (I-J^b), it behaves with the genetic restriction originally used to define the I-J locus.

It is possible that the I-E molecule of B26 has an alteration in its glycosylation pattern. It has been reported that Ia molecules on B cells and macrophages have different glycosylation patterns and that helper T cells can recognize these

Table 2. B26 can present antigen to T-cell hybridomas

Macrophage line	No. macrophages per well	[³ H]Thymidine incorporation, cpm		
		3D026.1	3D062.1	D011.10
SC8	2 × 10 ³	7,212 ± 881	7286 ± 1329	11,198 ± 982
B26	2 × 10 ³	10,623 ± 547	7610*	13,012 ± 3,923
SC8	1 × 10 ³	6,352 ± 1,996	3862 ± 520	10,430 ± 967
B26	1 × 10 ³	8,361 ± 1,053	5331 ± 869	10,450 ± 693

Data shown are averages of triplicate cultures ± SD. T-cell lines alone yielded 1953 ± 579, 1099 ± 865, and 1019 ± 313 cpm, respectively. The specificities of the T-cell hybridomas are 3D026.1, I-E^d and ovalbumin; 3D062.1, I-E^d; D011.10, I-A^d and ovalbumin.

*Single sample only.

Table 3. B26 activates suppressor T cells of the B10.A(5R) mouse but not of the B10.A(3R) mouse

Cells	Poly(Glu ⁵⁰ Tyr ⁵⁰)-TsF1	Poly(Glu ⁵⁰ Tyr ⁵⁰)-specific plaque-forming cells per culture	
		B10.A(5R) (I-J ^k)	B10.A(3R) (I-J ^b)
SC8 (H-2 ^{d/f})	—	1020 ± 600	1560 ± 340
	+	1800 ± 480	1740 ± 640
63 (H-2 ^{d/k})	—	2240 ± 360	1240 ± 280
	+	0 ± 120	2040 ± 160
B26 (H-2 ^{d/f})	—	1400 ± 420	3360 ± 380
	+	0 ± 740	2400 ± 20

The details are as described in the legend to Table 1.

differences (25). Dendritic cells contain less sialic acid than macrophages, and this property is thought to account for their efficiency as antigen-presenting cells (26). Furthermore, the difference between the B10.A(5R) and B10.A(3R) mouse strains has been hypothesized to be a glycosyltransferase (27). Although many glycosylation mutations are recessive, some are dominant and *in vitro* glycosylation mutants have been isolated from polyploid cell lines (28, 29). Notably Thy-1 mutants and T-cell receptor mutants of lymphoid cells have been shown to be glycosylation mutants. Recently a glycosylation mutant of a class I molecule was described; the mutation leads to decreased surface expression of class I (30). B26 on the other hand has the same level of membrane I-E^d expression as its parental line, SC8. If an I-E molecule altered with respect to glycosylation pattern or in other posttranslational modifications causes the change in haplotype restriction, the change must occur in regions that do not alter the interaction with helper T cells, as B26 retains its ability to present antigen to H-2^d-restricted helper T cells. This may imply that suppressor T cells and helper T cells recognize different epitopes on I-E molecules.

An alternative explanation is the presence of another molecule on macrophages, other than I-E, that is involved in the interaction with suppressor T cells. An adhesion molecule, I-CAM, on antigen-presenting cells has been shown to enhance antigen-presenting function to helper T cells (31). Some investigators have reported a peptide-binding molecule on antigen-presenting cell membranes that is closely associated with class II molecules and is involved in presenting antigen to T cells (32). It is not known whether other molecules besides I-E are involved in the interaction of antigen-presenting cells with suppressor T cells. To investigate this, we generated B26-specific antibodies that can inhibit the induction of poly(Glu⁵⁰Tyr⁵⁰)-specific suppression in the H-2^k haplotype by either splenic antigen-presenting cells or B26 cells but have no effect on the B26-mediated activation of H-2^d-restricted helper T cells. Preliminary immunoprecipitation experiments have not revealed the molecule recognized by these antibodies. We believe these antibodies recognize a determinant seen by poly(Glu⁵⁰Tyr⁵⁰)-

Table 4. B26 does not activate H-2^k-restricted helper T-cell clone 2H10

Macrophage line	³ H]Thymidine incorporation into CTLL-1 cells, cpm	
	+ Antigen	– Antigen
63 (H-2 ^{k/d})	19,259 ± 386	576 ± 488
B26 (H-2 ^{d/f})	327 ± 124	382 ± 43

B26 was assayed for its ability to activate the cytochrome c-specific I-E^k-restricted T-cell line 2H10. Data shown are averages of triplicate samples ± SD. Supernatant from the T-cell clone alone gave 424 ± 108 cpm.

Table 5. B26-specific antibodies block activation of suppression

Blocking* antibody	Poly-(Glu ⁵⁰ -Tyr ⁵⁰)-TsF1	Poly(Glu ⁵⁰ Tyr ⁵⁰)-specific plaque-forming cells per culture		
		BALB/k (H-2 ^k)	Splenic APC	BALB/c (H-2 ^d) SC8 as APC
None	—	1700 ± 40	1960 ± 280	1620 ± 100
	+	0 ± 260	0 ± 160	0 ± 240
2E7	+	1440 ± 180	1980 ± 280	0 ± 380
2F6	+	1660 ± 240	1380 ± 240	0 ± 60
Control				
IgG	+	0 ± 100	0 ± 60	0 ± 280

The details are as described in the legend to Table 1.

*Antibodies were used at 10 µg/ml during a 30-min incubation prior to the addition of poly(Glu⁵⁰Tyr⁵⁰)-TsF1. APC, antigen-presenting cells.

specific suppressor T cells of the H-2^k haplotype and this should be helpful in identifying the molecule on antigen-presenting cells important for haplotype restriction in the activation of at least some suppressor T cells.

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