IMMUNE RESPONSIVENESS OF SM/J MICE

Cellular Characteristics and Genetic Analysis of Hyperresponsiveness to B Cell Mitogens*

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B cell responses to mitogens or polyclonal B cell activators have been extensively studied because it is believed that such responses may provide valuable basic information regarding B cell differentiation (1), B cell subsets (2, 3), B cell functions (4, 5), and roles of B cells in autoimmune disease (6–8). There is some evidence that responsiveness to B cell mitogens is genetically determined. However, unlike T cell-dependent immune responses (9), B cell responses to mitogens in the mouse do not appear to be under control of genes located in the major histocompatibility complex. For example, the inbred strains of mice C3H/HeJ (10) and CBA/N (11) are unresponsive or poorly responsive to bacterial lipopolysaccharide (LPS),[1] a well-known B cell mitogen. The C3H/HeJ defect has been mapped to a single autosomal locus that is not linked to the *H-2* region (12), whereas the CBA/N defect is known to be due to an X-linked recessive gene (11, 13).

To further elucidate the genetic factors regulating B cell responsiveness to mitogens, we have tested splenic lymphocytes from a panel of inbred strains of mice for their proliferative responsiveness to a B cell mitogen derived from *Actinomyces viscosus* (AVIS). In the course of these screening experiments, we discovered that spleen cells from the SM/J strain exhibited dramatically high proliferative responses to AVIS mitogen. Further experiments disclosed that SM/J mice are hyperresponsive to LPS and other B cell mitogens as well. The studies reported here establish that the high proliferative response to mitogens is under polygenic, non-*H-2*-linked, autosomal genetic control. Furthermore, this genetic control appears to be expressed through the B cell because T lymphocytes or macrophages do not affect the high proliferative responses in vitro. In a future, related report,[2] we describe the significantly elevated natural killer cell activity in SM/J mice and its relationship to hyperresponsiveness of B cells to mitogens.

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[1] *Abbreviations used in this paper:* AVIS, a B cell mitogen from *Actinomyces viscosus*; B6, C57BL/6J mice; DxS, dextran sulfate; FCS, fetal calf serum; LPS, bacterial lipopolysaccharide; PBA, polyclonal B cell activator; PHA, phytohemagglutinin; PPD, purified protein derivative of tuberculin.

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Materials and Methods

Mice. Mice used for initial screening experiments were obtained from The Jackson Laboratory, Bar Harbor, Maine, and were between 8 and 20 wk of age. The SM/J and C57BL/6J (B6) mice used in subsequent studies were bred in our colony by brother-sister mating from breeding stock obtained from The Jackson Laboratory. The F_1 progeny of B6 female and SM/J male mice and the backcross progeny of (B6 × SM)F_1 females and B6 males were bred in our colony.

Mitogens. A crude homogenate from *A. viscosus* bacteria has prevously been shown to be a potent B cell mitogen in mice (14). For these experiments, we prepared a soluble cell-wall extract of the homogenate, which contains ~90% of the mitogenic activity, using a modification of the trichloroacetic acid extraction method of Schleifer and Kandler (15), as previously described (16).

LPS from *Escherichia coli* 055:B5 (Westphal extract) and phytohemagglutinin P (PHA) were obtained from Difco Laboratories, Detroit, Mich. Purified protein derivative of tuberculin (PPD) was obtained from the National Institute of Allergy and Infectious Diseases, Bethesda, Md. Dextran sulfate (DxS), type 500-S, was obtained from Sigma Chemical Co., St. Louis, Mo.

Lymphocyte Preparation and Culture Conditions. Spleen cells were prepared as previously described (14) and were suspended in RPMI 1640 medium containing penicillin (100 U/ml), streptomycin (100 μg/ml), 4 mM Hepes buffer, and 5% heat-inactivated normal human serum. Cells were dispensed in 0.1-ml vol into the wells of microtest plates (3040; Falcon Labware, Div. of Becton, Dickinson & Co., Oxnard, Calif.). Mitogens were suspended in medium at appropriate concentrations and added to the wells in 0.1-ml vol at the start of the cultures. The cells were cultured for 48 h at 37°C in a humidified atmosphere at 5% CO_2 in air. DNA synthesis was measured as described elsewhere (16). In previous experiments we demonstrated that optimal DNA-synthesis responsiveness of C57BL/6J mouse splenocytes to AVIS and LPS mitogens occurs on the 2nd d of culture (14). Optimal mitogen dosage was found in those experiments to be 100 μg/ml of AVIS or of LPS. Therefore, in the screening experiments reported here, we used 2 d of culture and a mitogen dosage of 100 μg/ml. We subsequently verified that the mitogenic response of SM/J cells was also greatest on day 2 of culture and that optimal mitogen doses were similar for SM/J and B6 mice.

Removal of B Cells. B lymphocytes were removed from splenocyte preparations by three different techniques: (*a*) antibody-complement lysis: cell surface immunoglobulin-positive cells were lysed by incubating 10^7 splenocytes with 1 ml of a 1:15 dilution of goat anti-mouse Ig for 45 min at 4°C. The cells were washed and incubated 30 min at 37°C at a concentration of 10^6 cells/ml with a 1:10 dilution of rabbit complement (Low-Tox-M; Cedarlane Laboratories, Hornby, Ontario, Canada). (*b*) Nylon wool column: B lymphocytes were removed from splenocyte preparations by two passages through nylon wool columns, as previously described (17). (*c*) Rosetting: B cells were selectively removed by rosetting with sheep erythrocytes coupled with affinity-purified antibody to mouse Ig using the technique of Walker (18).

Removal of T Cells and Macrophages. T lymphocytes were removed from splenocyte preparations using rabbit anti-rat brain antiserum and complement (19). Macrophages were removed by adherence to petri dishes for 1.5 h at 37°C using medium containing 15% fetal calf serum (FCS). The nonadherent cells were withdrawn from the dish, washed twice, and resuspended in medium containing 5% normal human serum. As a second method of eliminating macrophages, silica powder was added in a concentration of 50 μg per microculture, as previously described (20). Silica is selectively toxic for macrophages (21).

Immunofluorescence. The percentage of B cells present in splenocyte preparations was determined by immunofluorescent staining for cell surface immunoglobulin. Approximately 10^6 viable cells in 50 μl of serum-free medium were incubated with 25 μl of either fluorescein-conjugated, affinity-purified Fab′ fragments of a goat anti-mouse Fab antiserum or with rhodamine-conjugated $(Fab')_2$ goat anti-mouse IgM (heavy chain specific; N. L. Cappel Laboratories, Cochranville, Pa.) for 30 min. T cells were detected by staining in the same manner with fluorescein-conjugated Fab′ fragments of rabbit anti-rat brain antibody. The cells were counted under the fluorescence microscope as previously described (22).

H-2 Typing. Mouse alloantisera against $H\text{-}2^b$ and $H\text{-}2^v$ private specificities were obtained from the Research Reference Reagents Branch of the National Institute of Allergy and

Infectious Diseases, Bethesda, Md. *H-2* typing of spleen cells of (SM × B6)F_1 × B6 backcross progeny and parental and F_1 controls was done using a two-step cytotoxicity technique with specific alloantisera and a 1:15 dilution of rabbit complement (Low-Tox-H, Cedarlane Laboratories).

Results

Hyperresponsiveness of SM/J Splenocytes to AVIS and Other B Cell Mitogens. These studies began as a screening experiment in which two mice from each of several different strains were tested for splenocyte proliferative responsiveness to AVIS, a B cell mitogen. As a positive control and for comparative purposes, each assay also included cultures of splenocytes from B6 mice because the AVIS-induced response of these animals had previously been thoroughly characterized (14). The results of the screening experiment are shown in Fig. 1. No strains were unresponsive; although there were minor variations in the magnitude of response in individual mice, most strains did not differ remarkably from each other or from the control B6 mice. In sharp contrast was the very high response of the SM/J strain. The proliferative response of splenocytes from SM/J mice was found to be approximately five- to eightfold higher than the responses of all the other strains tested.

We next tested the proliferative responses of SM/J splenocytes to LPS, DxS, and PPD to distinguish whether the high response of SM/J was restricted to AVIS, or whether SM/J was a higher responder to several B cell mitogens. As shown in Table I, the responses to all of the B cell mitogens that we tested were also severalfold higher than the responses of B6 splenocytes to the same mitogens. It is important to note that

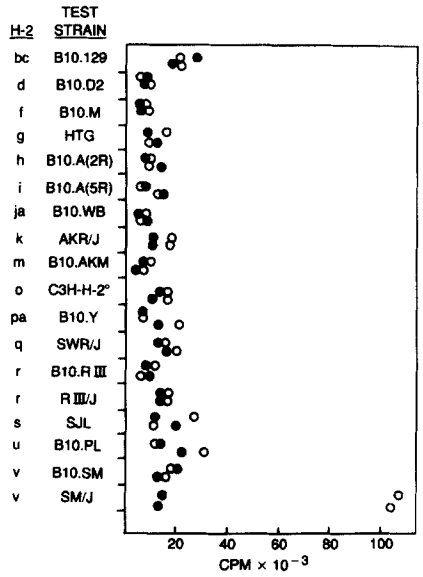

FIG. 1. The AVIS-induced proliferative responsiveness of splenocytes from a panel of inbred mouse strains. 4×10^5 cells/well were cultured for 48 h in the presence or absence of 100 μg/ml of AVIS mitogen. The cells received a pulse of ^{125}I-deoxyuridine 4 h before harvest. The data represent the mean counts per minute (cpm) of six replicate AVIS-stimulated cultures minus the mean cpm of six replicate nonstimulated cultures. Nonstimulated cultures were generally in the range of 200–1,200 mean cpm. Each point represents the data from one animal. Symbols: ○, test strain; ●, C57BL/6J control.

TABLE I

Comparison of the SM/J and C57BL/6J Responses to Several B Cell Mitogens and to PHA

Experiment	Mitogen‡	DNA synthesis*	
		SM/J	C57BL/6J
1	Medium control	845 (133)	1, 382 (282)
	AVIS	159, 113 (4,018)	7, 098 (580)
	LPS	160, 518 (14,596)	12, 660 (2,528)
	PHA	16, 187 (1,765)	21, 397 (2,525)
2	Medium control	1, 898 (271)	223 (49)
	AVIS	79, 861 (7,811)	9, 841 (1,582)
	DxS	17, 977 (1,867)	1, 299 (154)
	PPD	48, 870 (5,822)	6, 492 (1,516)

* Cultures containing 4×10^5 splenocytes per microtest well were pulsed for the final 4 h of 48-h culture periods with 0.5 μCi of ^{125}IUdR. The data represent the mean cpm (SD) of radiolabel incorporated by six replicate cultures.

‡ Mitogen concentrations: AVIS and LPS, 100 μg/ml; PHA, 10 μg/ml; DxS, 50 μg/ml; PPD, 100 μg/ml.

the response of SM/J splenocytes to PHA, a T cell mitogen, was not higher than that of B6 splenocytes. This finding of normal PHA responsiveness is in agreement with a previous report (23).

Genetic Analysis. No sex-linked differences in B cell responsiveness were evident in SM/J mice because both male and female mice were found to be hyperresponsive to B cell mitogens. The responsiveness to AVIS and LPS of (B6 × SM)F_1 progeny were compared to age- and sex-matched B6 and SM/J parental mice; the results of two such experiments, using AVIS mitogen, are shown in experiments 1 and 2 of Fig. 2. The F_1 mice consistently exhibited a mitogenic response to AVIS intermediate in magnitude compared with the responses of the parental strains. The LPS-induced responses were also intermediate (data not shown). These findings demonstrate that hyperresponsiveness in SM/J mice is genetically determined. The observed intermediate responsiveness of the F_1 mice is consistent with a gene-dosage effect, with semidominant expression of the SM high-response genes.

The B10.SM congenic-resistant strain that carries the SM/J $H\text{-}2^v$ haplotype is not a high responder strain to B cell mitogens (Fig. 1). This suggests that the genes conferring high responsiveness in SM/J mice are not present on the segment of chromosome 17 transferred to B10.SM. To definitively test *H-2* linkage of the high response genes, (B6 × SM)F_1 × B6 backcross progeny mice were *H-2* typed and their splenocyte proliferative responsiveness to AVIS and LPS was determined. Proliferative responsiveness to AVIS by splenocytes from backcross mice was compared with SM/J, (B6 × SM)F_1, and B6 mice. The results are shown in experiments 3–7 of Fig. 2. In general, the magnitude of the AVIS responses of backcross mice was intermediate to that of the F_1 and the B6 mice. Similar results were observed using LPS as a B cell mitogen (data not shown). The results indicate that the high responsiveness of SM/J is under polygenic control; backcross mice did not segregate into two discrete responder groups ("high" and "low") as would be expected for single gene control. Only 1 of the 36 backcross progeny tested had a response similar to the F_1 parental mice. Comparison of the mitogenic responses of $H\text{-}2^{b/v}$ and $H\text{-}2^{b/b}$ backcross mice

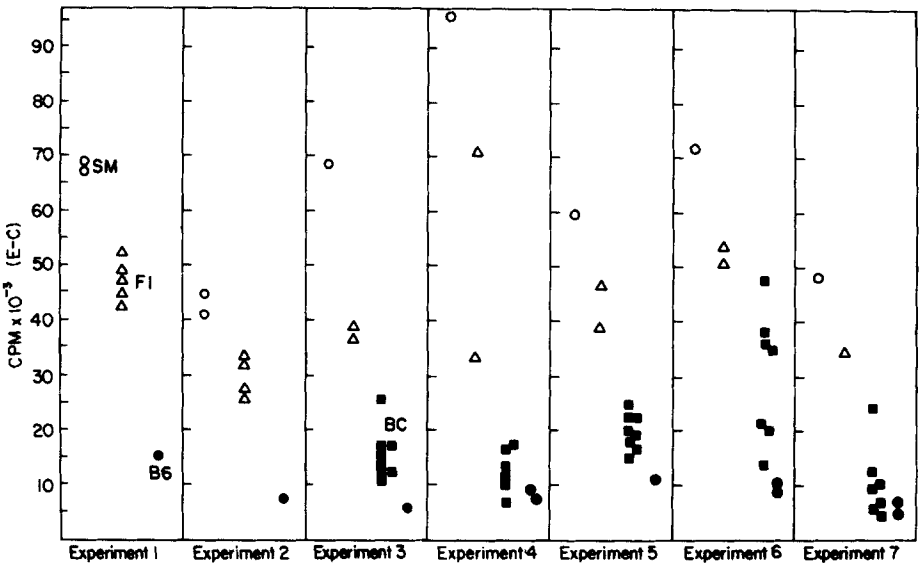

FIG. 2. Analysis of the AVIS-induced proliferative responsiveness of splenocytes from (B6 × SM)F_1 and (B6 × SM)F_1 × B6 backcross mice. Culture conditions were the same as described in Fig. 1. The data represent the mean cpm of six replicate AVIS-stimulated cultures minus the mean cpm of six replicate nonstimulated cultures. Nonstimulated cultures were generally in the range of 200–1,200 mean cpm. Experiments 1 and 2 compare the (B6 × SM)F_1 responses with the responses of SM/J and B6 parental strains. Experiments 3–7 compare the responses of backcross mice with the responses of SM/J, F_1, and B6 mice. Symbols: ○, SM/J; △, (B6 × SM)F_1; ■, (B6 × SM)F_1 × B6 backcross; ●, B6.

TABLE II

Lack of Association between H-2 Type and Mitogenic Responses in (B6 × SM)F_1 × B6 Backcross Mice

H-2 genotype‡	Number of mice	DNA synthesis*	
		AVIS§	LPS§
b/b	20	18,741 (2,268)	31,189 (3,121)
v/b	16	17,478 (2,438)	30,073 (3,571)

* Splenocytes were cultured as described in Table I, and DNA synthesis was quantitated by incorporation of a pulse of ^{125}IUdR. Data are expressed as the mean cpm (SEM).

‡ Genotypes were determined by a cytotoxicity assay using specific antisera to K and D alloantigens and rabbit complement.

§ AVIS and LPS mitogens were used at 100 μg/ml.

confirmed that the high responsiveness to B cell mitogens was not *H-2* linked (Table II). There were no differences in the mean responses or range of responses of these two groups.

High Responsiveness Is Not T Cell or Macrophage Dependent. Although earlier studies using B6 mice had shown the AVIS-induced proliferative response of splenocytes to be T independent (14), it was possible that T cells in the SM/J strain may have been participating in some way, such as by providing an enhancing signal. Therefore, T cells were selectively eliminated from SM/J splenocyte suspensions by lysis with antibody and complement, and the response of such T-depleted cultures was compared

with the response of intact SM/J splenocytes. As shown in experiment 1 of Table III, T cell depletion did not reduce the hyperresponse to AVIS, suggesting that T cells are not required.

The role of macrophages in proliferative responses of AVIS and LPS B cell mitogens, although negligible in CBA/J and C57B6/J mice (J. Clagett, K. Smith, and C. Rosse, manuscript in preparation), remained a possibility in SM/J mice. We therefore did experiments in which macrophages were removed by either adherence to plastic or poisoning with silica. The results of these experiments (Table III; experiments 2 and 3) illustrate that hyperresponsiveness to AVIS or LPS in SM/J mice is not reduced by removal of macrophages.

Removal of B Cells Abrogates Hyperresponsiveness. The experiments above support the hypothesis that hyperresponsiveness of SM/J splenocytes is due to an abnormality in the B cell population rather than to an unusual T cell- or macrophage-mediated enhancement of the B cell response. To definitively prove that the high response was due only to B cell proliferation, B lymphocytes were selectively removed from SM/J splenocyte suspensions by antibody-complement lysis, nylon wool, or rosetting techniques before mitogen stimulation. The results are shown in Table IV. All three methods used to remove B cells (antibody and complement lysis, passage of spleen cells through nylon columns, and rosetting B cells with αIg-coated sheep erythrocytes) effectively abrogated responsiveness to LPS. It is unlikely, therefore, that SM/J T cells or macrophages are proliferating in response to B mitogens; rather the observed high DNA synthesis response of SM/J splenocytes is due to elevated B cell proliferation. This was confirmed by immunofluorescent and electron microscopic examination of cells from intact SM/J splenocyte cultures 2 and 4 d after LPS stimulation. The vast majority of such cells (~80%) were immunoglobulin-producing cells with the fine structural features of B blasts or plasma cells. We did not observe any cells with T blast morphology.

Percentages of B and T Cells in SM/J Spleens. To determine whether the high

TABLE III

Failure of Removal of T Cells or Macrophages to Abrogate High Response of SM/J Splenocytes to B Cell Mitogens

Experiment	Method	Cells	DNA synthesis* Medium control	AVIS	LPS	PHA	Con A
1	RARB + C‡	Intact	839 (102)	74,390 (5,496)	ND§	11,763 (1,741)	ND
		T Cells removed	319 (50)	100,120 (10,996)	ND	335 (92)	ND
2	Adherence‖	Intact	4,302 (352)	47,681 (7,813)	103,381 (4,270)	ND	43,081 (5,082)
		Mϕ removed	3,715 (436)	54,670 (4,811)	92,739 (7,043)	ND	25,382 (3,883)
3	Silica¶	Intact	521 (140)	ND	42,762 (4,835)	20,044 (5,723)	122,671 (12,939)
		Mϕ removed	527 (151)	ND	51,145 (3,278)	22,339 (8,450)	91,674 (7,729)

* Culture and radiolabeling conditions were as described in Table I. Mitogen concentrations: AVIS and LPS, 100 μg/ml; PHA, 10 μg/ml; concanavalin A (Con A), 2.5 μg/ml. The data are presented as mean cpm (SD) of radiolabel incorporated by six replicate cultures.

‡ T cells were removed by treatment of the splenocytes with rabbit anti-rat brain (RARB) and complement (19). The remaining cells were washed and resuspended at the same concentration as control (untreated) splenocytes.

§ ND, not determined.

‖ Macrophages (Mϕ) were removed by preincubation of splenocytes on plastic petri dishes for 1.5 h at 37°C in medium containing 15% FCS. The nonadherent cells, containing <3% phagocytes, were collected and resuspended at the same concentration as control cells in medium containing 5% normal human serum.

¶ Mϕ were killed by addition of 50 μg of silica per microculture (20).

TABLE IV

Abrogation of High Response of SM/J Splenocytes to LPS by Removal of B Cells

Experiment	Method	DNA synthesis*				
		Cells	Medium control	LPS	PHA	Con A
1	αIg + C‡	Intact	936 (227)	96,370 (4,116)	ND§	266,400 (56,700)
		B cells removed	681 (208)	16,189 (2,380)	ND	213,973 (74,820)
2	Nylon wool‖	Intact	253 (46)	47,801 (8,497)	4,413 (672)	78,408 (10,073)
		B cells removed	117 (49)	516 (68)	15,303 (908)	28,150 (4,617)
3	Rosette¶	Intact	378 (45)	55,551 (4,547)	ND	85,743 (5,313)
		B cells removed	509 (119)	3,142 (182)	ND	68,905 (7,757)

* Culture and radiolabeling conditions were as described in Table I. Mitogen concentrations: LPS, 100 μg/ml; PHA, 10 μg/ml; concanavalin A (Con A), 2.5 μg/ml. The data are presented as mean cpm (SD) of radiolabel incorporated by six replicate cultures.

‡ B cells were lysed by treatment with goat-anti-mouse Ig (light chain specific) and rabbit complement. The remaining cells were washed and resuspended at the same concentration as control (intact) splenocytes.

§ ND, not determined.

‖ Splenocytes were passed twice over a nylon wool column to remove B cells (17).

¶ B cells were selectively removed by rosetting with erythrocytes coupled with antibody to mouse Ig (18).

TABLE V

Comparison of the Percentages of B and T Cells in Spleens of SM/J and C57BL/6J Mice

Fluorescent reagent*	Percentage of surface-stained cells‡	
	SM/J	C57BL/6J
Anti-Fab	51.6 (6.9)	55.8 (6.8)
Anti-μ	48.6 (1.7)	52.2 (2.9)
Anti-T cell (RARB)	28.9 (3.5)	27.4 (3.4)

* B cells were detected by staining viable cells with either affinity-purified anti-Fab antibodies or anti-μ specific antiserum. T cells were detected by staining viable cells with rabbit anti-rat brain (RARB) antiserum (19).

‡ The data are the mean values (SD) from three independent experiments. All mice were 8 wk of age.

mitogenic response was due to a higher than normal ratio of B cells to T cells in the SM/J spleen, we compared the cell surface phenotypes of SM/J and B6 splenocytes. Splenocytes from mice of each strain were stained with fluoresceinated antibodies to B and T cell markers and were examined by fluorescence microscopy. The results show that the percentages of splenic B and T cells were not significantly different in the two strains of mice (Table V).

Discussion

Our studies show that splenic B lymphocytes from SM/J mice undergo an unusually high proliferative response to several polyclonal B cell activators. It is unlikely that T

cells or macrophages play an enhancing role in this high response because their depletion from cultures does not alter the magnitude of response (Table III). We must, however, add the caveat that very small numbers of T cells or macrophages may be present in depleted cultures and that these few cells may contribute something essential to the high response. We do not believe that high responsiveness is due to a lack of, or reduced numbers of, suppressor T cells because the addition of varying numbers of splenocytes from normal responder B10.SM mice (which presumably have normal suppressor T cells) to cultures of SM/J splenocytes does not reduce the magnitude of the mitogenic response (unpublished results). We cannot rule out the possibility that SM/J B cells lack a functional receptor that is necessary for recognition of a regulatory signal. However, this too seems unlikely because the mice do not have hypergammaglobulinemia, leukocytosis, splenomegaly, or any apparent autoimmune diseases (see below).

Hyperresponsiveness to B cell mitogens in SM/J mice is genetically determined. The responses of (B6 × SM)F_1 mice to LPS and AVIS were intermediate compared with SM/J and B6 parents (Fig. 2); this suggests that a gene dosage effect is operative in the F_1 mice (expression at 50% of SM/J levels of a critical enzyme or receptor). Analysis of individual segregating (B6 × SM)F_1 × B6 backcross progeny indicates that hyperresponsiveness to B cell mitogens is under polygenic control in SM/J mice and that these genes are not linked to the *H-2* complex (Table II). However, the lack of a role for *H-2* in hyperresponsiveness of SM/J mice to the B cell mitogens LPS and AVIS does not preclude the possibility that *H-2*-linked genes may influence the response to other B cell mitogens in other strains of mice. Relatively little is known about genes controlling responsiveness to B lymphocyte mitogens. The best characterized genes that influence LPS responsiveness are the *Lps*^{d} gene (12) of the C3H/HeJ strain and the *xid* gene of the CBA/N strain (11, 13). We are presently developing and testing F_1 hybrids between C3H/HeJ, CBA/N, and SM/J to determine the influence of the *Lps*^{d} and *xid* genes on the expression of the SM/J hyperresponse genes.

B lymphocytes are heterogeneous, and there is evidence based upon sequential stimulation with various different mitogens and upon suicide experiments that subsets of B cells respond optimally to different B cell mitogens (24–26). However, Andersson et al. (27) have postulated that there may also be a B cell subset that expresses all mitogen receptors of a species and therefore responds to all mitogens. Because SM/J mice are hyperresponsive to four different B cell mitogens, which putatively activate different B cell subsets, there are at least three possible models to explain SM/J hyperresponsiveness: (*a*) a general expansion of all B cell subsets that respond to mitogens; (*b*) specific expansion of the B cell subset that has multiple-mitogen reactivity (27); (*c*) all subsets are normal in proportion, but the cell cycle of B cells in response to mitogens is shortened so that multiple rounds of DNA synthesis occur. These possibilities may be tested by using the 5-bromo-deoxyuridine suicide technique as described by Bona et al. (26) and by cell-cycle analysis using flow cytometry techniques (28).

SM/J mice provide a unique and potentially useful model system for testing a variety of questions regarding B cell triggering, differentiation, and function. In contrast to other strains with high spontaneous polyclonal B cell responsiveness—such as the motheaten (29–30), NZB, and MRL/1 strains (31)—SM/J mice are healthy

and have normal life spans with no detectable autoimmune disease processes. It has been reported that natural IgM thymocytotoxic antibodies are markedly elevated in SM/J mice, particularly in old females; however, SM/J mice do not have antinuclear antibodies or elevated serum IgM levels, and they do not develop systemic lupus erythematosus-like syndrome (32). We have screened several animals for immune complex deposition in the kidney and have found no evidence of this symptom of autoimmune disease. Serum levels of IgG are also similar for SM/J and B6 mice and do not increase significantly in aged SM/J animals. Furthermore, we have examined the kidneys of 10 aged mice (10–17 mo old) for amyloid by Congo red staining, and, in contrast to a previous anecdotal report (33), we have not found any evidence of amyloidosis. The lack of autoimmunity in SM/J mice suggests that in spite of hyperresponsiveness to B cell mitogens in vitro, they do possess an effective immunoregulatory system in vivo.

Summary

We tested the proliferative responses of splenocytes from a panel of inbred mouse strains to AVIS, a B cell mitogen from *Actinomyces viscosus* bacteria. The SM/J strain was found to exhibit severalfold higher responsiveness than any of the other strains. SM/J splenocytes were also hyperresponsive to the B cell mitogens lipopolysaccharide, dextran sulfate, and purified protein derivative of tuberculin, but responsiveness to the T cell mitogen phytohemagglutinin was normal. (B6 × SM)F_1 and F_1 × B6 backcross mice were tested for AVIS and lipopolysaccharide responsiveness, and it was determined that hyperresponsiveness was under polygenic, autosomal, non-*H-2*-linked gene control. Genetic control of response to B mitogens in SM/J mice appears to be expressed solely through the B lymphocyte because removal of T lymphocytes or macrophages did not reduce the magnitude of responsiveness in vitro. SM/J mice may provide a useful model for testing questions regarding B cell triggering, differentiation, and function, and to examine the genes involved with B cell proliferation.

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