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Delayed Clearance of Filarial Infection and Enhanced Th1 Immunity Due to Modulation of Macrophage APC Functions in xid Mice

Sangita Mukhopadhyay, Prakash Kumar Sahoo, Anna George, Vineeta Bal, Satyajit Rath, and Bala Ravindran

Bruton’s tyrosine kinase (Btk) mutant CBA/N mice show delayed clearance of injected microfilariae (mf) compared with wild-type CBA/J mice. Anti-mf T cells from CBA/N mice make relatively more IFN-γ than those from CBA/J mice. The anti-mf T cell proliferative responses are also greater in CBA/N mice. This CBA/N immune phenotype is not restricted to filarial Ags, because immunization with pure proteins also yields T cell responses of greater proliferative magnitude skewed away from Th2 cytokines in CBA/N compared with CBA/J mice. The increased magnitude of CBA/N T cell proliferative responses is reflected in increases in both precursor frequencies and clonal burst sizes of responding Ag-specific T cells, and is independent of the source of immunization with pure proteins also yielding T cell responses of greater proliferative magnitude skewed away from Th2 cytokines in CBA/N compared with CBA/J mice. The increased magnitude of CBA/N T cell proliferative responses is reflected in increases in both precursor frequencies and clonal burst sizes of responding Ag-specific T cells, and is independent of the source of re-stimulating APCs. Transfer of CBA/J peritoneal resident cells (PRCs) into CBA/N mice before pure protein immunization leads to a wild-type immune phenotype in the recipient CBA/N mice, with a reduction in the proliferative response and a relative decrease in the IFN-γ produced. When wild-type PRC subpopulations are similarly transferred, the wild-type immune phenotype is transferred by macrophages rather than by B cells. Transfer of wild-type PRCs into CBA/N mice before injection of mf also causes similar changes in the anti-mf T cell responses and enhances the clearance of mf. Thus, Btk is involved in critical macrophage APC functions regulating priming of T cells, and can modulate these responses in pathophysiologically relevant fashion in vivo. The Journal of Immunology, 1999, 163: 875–883.

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Abbreviations used in this paper: mf, microfilaria; Btk, Bruton’s tyrosine kinase; EIA, enzyme-linked immunosorbent assay; OA, chicken OVA; PRC, peritoneal resident cell; xid, X-linked immunodeficient.
Materials and Methods

Mice

Btk mutant X-linked immunodeficient (xid) CBA/N mice and their wild-type counterparts, CBA/J mice, obtained from The Jackson Laboratory (Bar Harbor, ME) and bred in the small animal facility of the National Institute of Immunology, New Delhi, India, were used for all experiments at 6–10 wk of age. All animal experimentation was done with the approval of the Institutional Animal Care and Usage Committees.

Parasites

Fresh adult-stage parasites of Setaria digitata were collected from the peritoneal cavities of abattoir-slaughtered cattle and live mf of S. digitata were obtained by dissection of mature female worms in vitro (48).

Microfilaremia in mf-infected mice was followed by biweekly counts of mf in thick smears from 20 μl of blood stained with Giemsa stain (3% for 1 h) (48).

Preparation of mf antigenic extracts

About 1 × 10^6 mf of S. digitata were crushed in ice-cold PBS and then sonicated (Sonifier, Branson, Danbury, CT) using 10 cycles each of 4 min. To prepare Ag for Ab detection assays, these sonicated preparations were centrifuged at 7000 rpm for 30 min, filtered through a 0.22-μm filter, and stored as aliquots at –70°C. For use in T cell cultures, the sonicated preparations were sterilized by autoclaving and stored at –70°C.

Immunization

Mice were infected intraperitoneally with 1–5 × 10^6 mf of S. digitata per animal and sacrificed at various days postinfection as indicated.

Chicken OVA (OA) was used either as a native Ag or after maleylation, which enables it to bind to scavenger receptors (49) and helps generate a positive T cell response of higher magnitude (50) and greater relative prominence of IFN-γ (21). Mice were immunized with 1 mg OA in PBS intraperitoneally and sacrificed on days 7–14 postimmunization.

Maleylation of proteins

Maleylation to acylate ε-amino groups on lysine residues in the proteins was conducted as described (49). Briefly, the native protein was dissolved in borate buffer of pH 8.5 at 10 mg/ml. Finely powdered maleic anhydride (Sigma, St. Louis, MO), 2.5-fold higher in quantity than the total protein amount, was added in with constant stirring keeping the pH between 8.5 and 9.0 with NaOH throughout. At the end of the reaction, the pH of the mixture was adjusted to 7.4, and excess maleic anhydride was removed by extensive dialysis against PBS at 4°C. The degree of maleylation was estimated using the trinitrobenzenesulfonic acid assay for free ε-amino groups (51), and maleyl proteins were used for experiments only when they showed >90% maleylation.

Ab assays

Enzyme-linked immunosorbent assays (ELISAs) were used to determine the Ag-specific IgM, IgG, IgG1, and IgG2a Abs in sera from immunized mice. Briefly, 96-well polystyrene microtiter plates (Nunc, Roskilde, Denmark) were coated with fluid-phase mf antigenic extract at 10 μg/ml overnight at 4°C. The plates were then washed and blocked with 1% nonfat milk protein. Serially diluted sera were then added, and bound Abs revealed, after washing, by goat anti-mouse IgG1 or IgG2a followed by HRP-coupled rabbit anti-goat Ig (Jackson ImmunoResearch, West Grove, PA), or goat anti-mouse IgG-biotin followed by streptavidin-HRP (Genzyme, Cambridge, MA) or by rabbit anti-mouse IgM-HRP (Jackson ImmunoResearch). HRP was detected using o-phenylenediamine tetrahydrochloride (Sigma) in citrate-phosphate buffer (pH 5.4) and H₂O₂ (Qualigen, Mumbai, India). The reaction was stopped using 1 N H₂SO₄, and the absorbance values were read at 492 nm (EL340, Bio-Tek, Burlington, VT). The concentrations of IgM and IgG Abs were calculated from standard curves generated with appropriate affinity-purified standards, and IgG1/ IgG2a ratios were calculated using reciprocal log titers at half-maximal absorbance.

Preparation of cellular subpopulations

Mice were sacrificed by cervical dislocation, and splenic cells were isolated. Where necessary, T cells were purified by loading spleen cell preparations onto 6-nl nylon wool columns at 1 × 10^6 per column and collecting the nonadherent cells. The purity of the T cell preparation so isolated was assessed by flow cytometrically by anti-CD3 staining, and the population was used when T cell purity was >95%. Splenic cells (2 × 10^6) from nonimmunized animals were used as APCs when purified T cells were used as responders.

Peritoneal resident cells (PRCs) were harvested by peritoneal lavage with chilled sterile tissue culture medium. Peritoneal cell types were separated by plating them (30 × 10^6 in 5 ml of medium) in 100-mm petri plates (Falcon, Franklin Lakes, NJ) and incubating at 37°C for 60 min. Nonadherent cells were then removed and the plates washed with medium. The adherent macrophages were detached by vigorous flushing with medium. B cells were purified from the nonadherent cells by panning on rabbit anti-mouse Ig-coated 100-mm petri plates (Falcon) at 37°C for 1 h, followed by washing. Adherent B cells were harvested by repeated flushing with medium. Cell preparations were analyzed by flow cytometry and used when >90% pure.

T cell activation assays

Cells were suspended in Click’s EHAA medium (Irvine Scientific, Santa Ana, CA) containing 0.05 mM 2-ME, 10% FCS (HyClone, Logan, UT), and antibiotics (100 U/ml of penicillin and 100 μg/ml of streptomycin). They were cultured (4 × 10^6/well) with titrated doses of Ags in a final volume of 100 μl of medium well at 37°C. After 60 h of incubation, 100 μl of supernatant per well was collected for estimation of cytokine levels. Cell proliferation was assayed by pulsing the cultures with 0.5 μCi [3H]thymidine (NEN, Boston, MA) per well at 96 h (48 h for mitogen-activated cultures) and incubating for an additional 10–12 h, and then harvesting and counting the plates on a scintillation counter (Betaplate, Pharmacia-Wallac, Turku, Finland). All assays were done in triplicates, and data expressed as mean cpm ± SE.

For limiting dilution analyses, graded numbers of purified responder T cells were plated into round bottom 96-well plates (Nunc; 48 wells per dilution) with 100 μg/ml of Ag and 3 × 10^5 naive splenic cell APCs per well. Control wells received no Ag. Cultures were pulsed with [3H]thymidine, harvested, and counted as above. Wells showing thymidine incorporation more than 2-fold higher than in negative control wells were scored positive. Ag-specific T cell frequencies were calculated by the minimum x² method (52). The mean cpm value of the positive-scoring wells at clonal frequencies was used as an estimate of the clonal burst size of the responding T cells.

Cytokine assays

Supernatants collected for cytokine assays were used for measurement of IL-5, IL-10, and IFN-γ by two-site sandwich ELISAs (Duoset, Genzyme; and Bio ricerca, San Diego, CA). In brief, plates were coated with monoclonal anti-IL-5, anti-IL-10 or anti-IFN-γ Ab and incubated overnight at 4°C. Following blocking with 1% BSA in PBS, 100 μl of culture supernatants were added to the plate and further incubated at 4°C overnight. This was followed by addition of the appropriate biotinylated polyclonal Abs at manufacturer-recommended concentrations, followed by streptavidin-HRP. Color was developed following the manufacturers’ protocols. Standard curves for the cytokines were obtained using the recombinant standard proteins provided by the manufacturers. Cytokine ratios were calculated at each recall Ag dose where necessary.

Statistical analysis

Statistical analysis was done using Student’s t test where appropriate.

Results

Btk mutant CBA/N mice show poor clearance of microfilaremia despite good Ab responses

There are previous reports that CBA/N mice are less capable of clearing microfilaremia than wild-type CBA/J mice (53, 54). We used the asexual blood stage of the cattle filarial parasite, S. digitata. One million mf per mouse were administered to CBA/J or CBA/N mice, and the persistence of mf in circulation was monitored over the succeeding 5–6 wk. Fig. 1A shows that CBA/N mice had significantly delayed mf clearance compared with CBA/J mice, so that by day 30 post-mf injection, no mf were detectable in peripheral blood in CBA/J mice, whereas the microfilaremia had only come down to about 400 mf/ml from a peak of 700 mf/ml of blood in CBA/N mice.

We examined the role of the immune system in this delayed mf clearance in CBA/N mice. Given the fact that B cell functions in CBA/N mice, so that by day 30 post-mf injection, no mf were detectable in peripheral blood in CBA/J mice, we began...
mated (mean 6 were determined (mean 6 anti-mf IgM and IgG (mean 6 mice. However, while the levels of IgG1 in the serum anti-mf responses were somewhat lower in CBA/N mice (0.05), their IgG responses were at least equivalent to those seen in CBA/J mice. However, while the levels of IgG1 in the serum anti-mf

CBA/N mice mount a higher and more Th1-dominated anti-mf immune response than wild-type mice

We then examined the Th1/Th2 cytokine balance directly in T cell cultures. The cytokine levels generated by mf Ag from splenic cells of infected mice at day 30 postinfection showed that T cells from CBA/N mice made more IFN-γ (Fig. 2C) and less IL-5 (Fig. 2A) than CBA/J mouse T cells did (although IL-10 levels were equivalent (Fig. 2B)). The IL-5/IFN-γ and IL-10/IFN-γ ratios were thus significantly different between the two strains (p < 0.01 and p < 0.001, respectively) with a relative Th1 bias seen in CBA/N mice (Fig. 2D).

T cell proliferative responses to S. digitata mf were also higher in mf-infected CBA/N mice at 60 days postinfection than in CBA/J mice (Fig. 2E), although proliferative responses to the T cell mitogen Con A were similar in the two strains while responses to a B cell mitogen, bacterial LPS, were somewhat lower in CBA/N mice (Fig. 2F) as expected (58).

CBA/N mice show T cell responses with relatively more IFN-γ than CBA/J mice against pure protein Ags

We next analyzed the T cell responses of CBA/J and CBA/N mice to immunization with pure protein Ags. Age-matched CBA/J and CBA/N mice were immunized, without adjuvant, with either native or maleyl-OA in PBS, and 7–10 days later the IFN-γ and IL-10 elicited by Ag from their splenic cells were analyzed. Fig. 3 shows that, regardless of whether the immunogen or the recall Ag used was native or maleyl-OA, CBA/N mice made less IL-10 (Fig. 3, A and C) and more IFN-γ (Fig. 3, B and D) than wild-type CBA/J mice did. Thus, the Th2/Th1 balances, as represented by the IL-10/IFN-γ ratios, were significantly (p < 0.01) lower in CBA/N mice than in CBA/J mice (Fig. 3E), no matter whether native or maleyl-OA were used for immunization or recall. The IL-5/IFN-γ ratio also shows a similar divergence between CBA/J and CBA/N mice as shown in another experiment where mice were immunized with native OA in PBS and recall assays were done 10 days postimmunization, and IL-5, IL-10, and IFN-γ levels elicited from their splenic cells by OA were quantitated. In addition to the enhancement of IFN-γ and decrease in IL-10 in CBA/N mice as already seen in Fig. 3, a decrease in IL-5 responses in CBA/N mice was also observed (Fig. 4A), with a resultant significant (p < 0.001) decrease in the IL-5/IFN-γ and IL-10/IFN-γ ratios in CBA/N mice (Fig. 4B).

CBA/N T cell responses are of larger magnitudes than CBA/J responses

In addition to the modulation of the Th1/Th2 balance in CBA/N mice, the T cell proliferative responses also show significant alteration. OA- or maleyl-OA-immune CBA/N mice showed significantly greater proliferative recall responses in vitro than CBA/J mice did (Fig. 5, A and B). Because B cell numbers may vary between xid and wild-type mice, purified splenic T cells from maleyl-OA-immune mice were tested for their anti-maleyl-OA pro-

FIGURE 1. S. digitata microfilaraemia and anti-mf Ab responses in CBA/N and CBA/J mice. CBA/J (C) and CBA/N (D) mice (6/group) were given S. digitata mf intraperitoneally and circulatory microfilaraemia was followed over time (mean ± SE; A). The data are representative of several experiments. On day 30, in both the CBA/J (C) and CBA/N mice (D), anti-mf IgM and IgG (mean ± SE; B) as well as anti-mf IgG1 and IgG2a were determined (mean ± SE; C, left axis) and IgG1/IgG2a ratios estimated (mean ± SE; C, right axis).
liferative responses in bulk assays, where they showed similar differences (data not shown). We then used these purified T cells to estimate the frequency of the Ag-specific T cells by titrating their number with fixed concentrations of APCs and Ag in limiting dilution assays. The enhanced responsiveness was seen to be the result of a substantial increase in the precursor frequency of Ag-specific T cells as shown by estimates from maleyl-OA-immune mice; against a CBA/J frequency of 1/3000, CBA/N mice had a frequency of 1/100 no matter which APCs were used (Fig. 5C). This assay also allowed an estimate of the ability of a single T cell to generate progeny from the clonal burst size as reflected by the average magnitude of the proliferative response in responding wells at clonal frequency (positive wells, <37%). There was a significant ($p < 0.01$) increase in the clonal burst size of the responding T cells from CBA/N mice (Fig. 5D). The increase in precursor frequency as well as in burst size in CBA/N mice was seen irrespective of whether the APCs used were of CBA/N or CBA/J origin (Fig. 5C).
CBA/J APC-mediated alteration of T cell responses and enhancement of microfilarial clearance in CBA/N mice

We next examined the possibility of alterations in the T cell priming functions of CBA/N APCs. We used an experimental system of adoptive transfer of wild-type CBA/J PRCs intraperitoneally into CBA/N mice just before i.p. immunization with maleyl-OA in PBS. Control CBA/J mice receiving CBA/J PRCs mounted proliferative (Fig. 6A), IL-10, and IL-10/IFN-γ responses that were essentially unaltered. However, if CBA/N mice had received CBA/J PRCs, they now showed reduced proliferative responses (Fig. 6A), as well as cytokine response alterations in the form of increases in IL-10 and reductions in IFN-γ levels (Fig. 6B and C), making their responses indistinguishable from those of CBA/J mice. These differences can be easily perceived in the Th2/Th1 balances as represented by IL-5/IFN-γ and IL-10/IFN-γ ratios (mean ± SD) for CBA/J (■) and CBA/N (□) mice.

In keeping with these data, repeated CBA/J PRC transfer into CBA/N mice after infection with S. digitata mf led to a faster clearance of the mf from peripheral circulation (Fig. 7A), confirming the crucial role of APCs. Expectedly, in CBA/N mice given CBA/J PRCs, anti-mf T cell responses are also modulated so that they resemble the CBA/J anti-mf response profiles. Thus, the proliferative T cell responses are reduced (Fig. 7B). Similarly, the levels of IL-5 and IL-10 are enhanced (Fig. 7, C and D) and the levels of IFN-γ reduced (Fig. 7E). Although these differences appear modest, they are reproducible in multiple experiments and lead to a significant (p < 0.05) alteration in the IL-5/IFN-γ and IL-10/IFN-γ ratios (Fig. 7F) and to faster mf clearance (Fig. 7A).

Identification of the cell type responsible for the T cell immune modulation seen in CBA/N mice

Because peritoneal APCs have both macrophages and B cells, we next transferred purified B cells and macrophages from CBA/J PRCs into CBA/N mice before immunization in a similar experiment. The results show that if CBA/N mice had received CBA/J B cells, they mounted proliferative (Fig. 8A), IL-5, IL-10, and IFN-γ responses (Fig. 8, B–D) that were essentially similar to those of control CBA/N mice. However, in CBA/N mice receiving CBA/J APRCs, the T cell immune modulation seen in CBA/N mice is preserved, suggesting that CBA/J B cells are the primary targets of CBA/J APCs.
PRC macrophages, the proliferative response was reduced (Fig. 8A), and the cytokine responses showed reduction in IFN-\(\gamma\) and increase in IL-5 and IL-10 levels (Fig. 8, B–D), leading to significant increases in the IL-5/IFN-\(\gamma\) and IL-10/IFN-\(\gamma\) ratios (mean \(\pm\) SE) in the two mouse strains for both control mice (■) and mice receiving CBA/J PRCs (□). Results shown are representative of three independent experiments.

Discussion

The data presented in this paper demonstrate that CBA/N mice mount a relatively more Th1-dominated T cell immune response of higher magnitude against filarial Ags (Fig. 2) as a consequence of altered macrophage function and yet clear S. digitata microfilaria more slowly than CBA/J mice do (Fig. 1), suggesting that Th1 responses may in fact be associated in some instances with susceptibility to filarial infections. Delayed clearance of microfilaraemia has been observed in xid mice using other model filarial parasites such as Brugia malayi (53) and Litomosoides sigmodontis (54). We have also observed that L3-stage infective larvae of the human filarial parasite B. malayi, which die soon after infection in CBA/J mice without maturation, survive better in CBA/N mice and develop to the juvenile adult stage (59), although they do not develop to the sexually mature adult stage as observed in xid mice for L. sigmodontis (54).

Our data that the Th2/Th1 balance is significantly shifted toward Th1 cytokines in CBA/N mice are consistent with findings from the L. sigmodontis-BALB/c mouse model, where depletion of CD4 T cells led to reduction in Th2 cytokines, eosinophilia, and IgE levels as well as to increased filarial susceptibility (60). The two systems are also consistent in finding deficiency in the Ab responses either of the IgM isotype (Fig. 1), or to T-independent parasite Ags such as phosphorylcholine (54) as expected from the known B cell defect in xid mice (46, 47). However, while a causal
correlation has been suggested between the reduced T-independent Ab responses and the lower IL-10 production from B cells in CBA/N mice as the result of the Btk mutation. To address this, we immunized the mice with pure protein Ags. We have shown previously that immunization with scavenger receptor-targeted protein Ags without adjuvant generates an immune response modulation seen in CBA/N mice. Immunization with native or maleylated chicken conalbumin (data not shown). Thus CBA/N mice have an intrinsic alteration in their immune response that is not dependent on the nature of the immunogen.

There are suggestions that Th1 immune responses may be involved in clearance of filariasis, based on the findings that antifilarial T cells from chronically microfilaraemic asymptomatic individuals produce less IFN-γ than do T cells from humans or monkeys with chronic microfilaraemic filariasis (10, 12, 61). However, data from a number of animal models have suggested that Th2 cytokines are associated with resistance to filarial infection (14–16). The previous data from the xid mouse model (54) as well as the present findings also support the argument that Th2 cytokines are associated with antifilarial resistance. It is interesting to note that the differences in the qualitative T cell responses between CBA/N and CBA/J mice we have observed are far more prominent in the cytokine ratios, reflecting the balance of the Th2/Th1 cytokines made than in the absolute levels of individual cytokines. Physiologically, it may be this balance that determines the outcome of infections, rather than isolated single cytokine-mediated effects.

It remains possible that, in addition to the Th2 cytokines, other factors in CBA/N mice may also contribute to the delayed mf clearance observed in them. The altered IgG1 to IgG2a ratios, caused by a marginal decrease in IgG1 and a pronounced increase in IgG2a (Fig. 1), and the different functional profiles of the two isotypes, may contribute to the delayed mf clearance in CBA/N mice. More importantly, the alteration in macrophage properties in the absence of Btk may extend to antiparasitic effector functions of macrophages, contributing to delayed mf clearance.

We have used IFN-γ as a typical Th1 cytokine, and IL-5 as well as IL-10 as Th2 cytokines, because IL-4 was not detectable in culture supernatants from these experiments (data not shown). Although IL-10 is somewhat more ambiguous than IL-4 in that it is also produced by cell types other than T cells such as macrophages (11) and B cells (54), the “background” levels of IL-10 in the absence of any recall Ag in these cultures were below the limits of detection, and it is thus likely that the IL-10 being detected upon antigenic stimulation here is in fact of T cell origin.

One question raised by the data using mf was whether the immune response modulation seen in CBA/N mice was a consequence of some combined influence of filarial parasites and Btk deficiency, or whether it was a common change in the immune properties of CBA/N mice as the result of the Btk mutation. To address this, we immunized the mice with pure protein Ags. We have shown previously that immunization with scavenger receptor-targeted protein Ags without adjuvant generates an immune response of greater proliferative magnitude (50) and a relative prominence of IFN-γ (21) than immunization with the native form of the Ag does, and it was therefore of interest to examine whether any modulation of the response magnitude and the T cell cytokine balance in CBA/N mice would hold good for both Th1-dominated and Th2-dominated responses.

Clearly, the immune deviation noted in CBA/N mice is not restricted to filarial Ags, because responses to pure protein immunization also led to relatively greater Th1-dominated T cell immune responses (Figs. 3 and 4) of higher magnitude (Fig. 5) in CBA/N than in CBA/J mice. This remained true whether native or maleylated OA were used, and in a situation where no adjuvant was used for immunization. Similar results were also obtained in immunizations with native or maleylated chicken conalbumin (data not shown). Thus CBA/N mice have an intrinsic alteration in their immune response that is not dependent on the nature of the immunogen.

In addition to the altered Th1/Th2 balances, this alteration also results in enhanced bulk proliferative Ag-specific T cell responses for which both increased precursor frequency and increased clonal...
burst size (Fig. 5) are contributory factors. However, mitogen-induced T cell responses are comparable (Fig. 2). These data suggest that Ag-induced commitment to a proliferation-competent secondary T cell is enhanced in CBA/N mice, implicating an APC function. Because CBA/N mice have a drastic reduction in the B-I B cell compartment in the peritoneum (62), it is of interest to note that their T cell hyperresponsiveness is not a finding restricted to intraperitoneal immunization, because anti-maleyl-OA proliferative responses of lymph node cells from CBA/J or CBA/N mice immunized with maleyl-OA in alum 14 days earlier showed similar differences (data not shown). However, the use of APCs from either strain evoked equivalent proliferative responses from primed T cells of either strain (Fig. 5).

Thus, while it is possible that the altered T cell responses seen in CBA/N mice could be due to intrinsic changes in T cell behavior, they are more likely to be due to changes in those costimulatory properties of CBA/N APCs that are important for T cell priming, such as through CD40, may be deficiently transduced by the absence of functional Btk, and in other experiments we have observed that inducible NO synthase induction is lower, leading to enhanced IL-12 production by xid macrophages. It is also possible that signals received by APCs in the course of T cell priming, such as through CD40, may be deficiently transduced by xid macrophages as is reported for xid B cells (66), contributing to the immune phenotype seen. The role of the Btk molecule in macrophage APC functions is thus likely to provide significant insights into a variety of pathways.

In conclusion, we have shown that Btk mutant CBA/N mice mount T cell responses of greater magnitude that are skewed towards production of IFN-γ as compared with wild-type CBA/J mice, and such a modulation of the T cell response is also seen in a model of infection with filarial parasites. This finding may contribute to the explanation of many of the phenotypes reported for the outcomes of a variety of infections in xid mice (67–69). This skewing of the T cell response in Btk mutant mice is the consequence of altered macrophage function, demonstrating a novel functional dimension of the Btk molecule. The Btk molecule has been shown to be involved in a wide range of signal transduction processes in B cells (70–72). The expression of Btk in myeloid cells and monocytes is well known (45). However, there are fewer data documenting its role in signaling in non-B cell populations, although mast cell signaling via Fce receptors has been shown to involve Btk (73, 74). The fact that Btk is involved in controlling some macrophage function crucial for T cell priming implies that Btk may be regulating the signal transduction pathway for some inducible costimulatory molecule.

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References


