

Hyperresponsive T_H2 cells with enhanced nuclear factor- κ B activation induce atopic dermatitis-like skin lesions in Nishiki-nezumi Cinnamon/Nagoya mice

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Background: Nishiki-nezumi Cinnamon/Nagoya (NC/Nga) mice raised in nonair-controlled conventional circumstances spontaneously develop atopic dermatitis-like skin lesions; however, the underlying mechanisms remain unclear. **Objective:** We wanted to identify the critical intracellular signaling molecules in T cells that induce atopic dermatitis-like skin lesions in NC/Nga mice. **Methods:** We examined the levels of signal transduction and cytokine production in T cells, particularly those in atopic dermatitis-like lesions induced by the topical injection of mite antigens in NC/Nga mice under specific pathogen-free conditions. **Results:** In NC/Nga mice maintained under specific pathogen-free conditions, the capability of T_H1/T_H2 and T cytotoxic 1/T cytotoxic 2 (Tc1/Tc2) cell differentiation increased significantly. T-cell antigen receptor-mediated activation of the extracellular signal-regulated kinase/mitogen-activated protein kinase cascade and nuclear factor- κ B (NF- κ B) signaling were enhanced in NC/Nga T cells. The expression of T_H2 cytokines (IL-4, IL-13, and IL-5) and that of GATA-binding protein 3 (GATA3), avian musculoaponeurotic fibrosarcoma (c-Maf), NF- κ B, and activator protein 1 (AP1) selectively increased in draining lymph node T cells of NC/Nga mice. Moreover, the cell transfer of inhibitory NF- κ B mutant-infected T_H2 cells reduced ear thickness in the mite antigen-induced skin lesion of NC/Nga mice.

Conclusion: Hyperresponsive T_H2 cells with an enhanced activity of NF- κ B and AP1 play a crucial role in the pathogenesis of atopic dermatitis-like skin lesions in NC/Nga mice. **Clinical implications:** These results indicate potential therapeutic usefulness of developing selective inhibitors for NF- κ B in the treatment of human atopic dermatitis. (J Allergy Clin Immunol ■■■■;■■■:■■■-■■■.)

Key words: Atopic dermatitis, NC/Nga mouse, NF- κ B, AP1, ERK/ MAPK cascade, T_H2

Atopic dermatitis (AD) is a multifactorial allergic inflammatory skin disorder characterized by pruritic chronic eczema, elevated serum IgE levels, and massive cellular infiltrates including T cells, eosinophils, mast cells, and macrophages.¹⁻³ Both genetic and environmental factors are involved in the pathogenesis of atopic diseases, including AD.⁴⁻⁶ In the acute phase of AD, T_H2-dependent inflammatory responses predominate, whereas the chronic eczematous phase that follows is dominated by the T_H0 and T_H1 responses with increased IFN- γ production.^{3,7,8} In addition, the involvement of type-2 cytokine secreting CD8 T cytotoxic (Tc) cells (Tc2 cells) in AD development is suggested.^{9,10}

The NC/Nga mouse is a reliable experimental model for human AD,¹¹ because the symptoms shown by Nishiki-nezumi Cinnamon/Nagoya (NC/Nga) mice are similar to those of human AD from a clinical, pathological, and immunologic point of view. When NC/Nga mice are maintained under air-uncontrolled conventional circumstances, the majority of mice spontaneously develop AD-like skin lesions with a marked elevation in the plasma levels of total IgE.¹² However, NC/Nga mice showed no clinical symptoms under specific pathogen-free (SPF) conditions. An immunohistochemical examination of the skin lesions in NC/Nga mice revealed the typical features of affected skin observed in patients with AD, such as an increased infiltration of T cells, mast cells, and eosinophils, and a substantial expression of IL-4 and IL-5.¹² A possible involvement of hyper-IgE production¹³ and a T_H2-biased status^{14,15} in the pathogenesis of AD-like skin lesions in NC/Nga mice have been reported. In addition, a

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Supported by grants from the Ministry of Education, Culture, Sports, Science and Technology (Japan); Grants-in-Aid for Scientific Research in Priority Areas #17016010 and #17047007, Scientific Research B #17390139, Scientific Research C #18590466, Grant-in-Aid for Young Scientists #17790318, and Special Coordination Funds for Promoting Science and Technology, the Ministry of Health, Labor and Welfare (Japan), the Japan Health Science Foundation, the Kanae Foundation, the Uehara Memorial Foundation, and the Mochida Foundation.

Disclosure of potential conflict of interest: The authors have received grant support from the Japanese government and foundations.

Received for publication December 26, 2005; revised May 16, 2006; accepted for publication May 23, 2006.

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0091-6749/\$32.00

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doi:10.1016/j.jaci.2006.05.024

Abbreviations used

AD:	Atopic dermatitis
DL:	Draining lymph node
Dp:	<i>Dermatophagoides pteronyssinus</i>
EMSA:	Electrophoretic mobility-shift assay
ERK:	Extracellular signal-regulated kinase
IκB:	Inhibitory nuclear factor-κB
IL-2R:	IL-2 receptor
IL-4R:	IL-4 receptor
MAPK:	Mitogen-activated protein kinase
NC/Nga:	Nishiki-nezumi Cinnamon/Nagoya
NFAT:	Nuclear factor of activated T cells
NF-κB:	Nuclear factor-κB
SPF:	Specific pathogen-free
STAT:	Signal transducer and activator of transcription
Tc:	T cytotoxic
TCR:	T-cell antigen receptor

dispensable role for signal transducer and activator of transcription (STAT) 6 and T_H2 cells has also been suggested.¹⁶ These variations in the results appears to be a result of possible differences in the experimental systems, where the dermatitis is induced spontaneously under conventional conditions, and the frequency and the severity of diseases are different at different facilities.

METHODS

Animals

NC/Nga mice¹¹ raised under SPF conditions were purchased from SLC (Shizuoka, Japan). C57BL/6 and BALB/c mice were purchased from Clea Japan (Osaka, Japan). All mice used in this study were maintained under SPF conditions. Animal care was performed in accordance with the guidelines of Chiba University.

Injection of mite antigens and depletion of CD4 or CD8 T cells

The mice were immunized with mite antigens as described previously.¹⁷ Briefly, NC/Nga mice were injected intradermally with saline or 5 μg *Dermatophagoides pteronyssinus* (Dp) extract (Cosmo Bio LSL, Tokyo, Japan) dissolved in 10 μL saline, on the ventral side of right ear. The mice were administered intraperitoneally with 100 μg anti-CD4 mAb (GK1.5), anti-CD8 mAb (53-6.7; BD Biosciences, San Jose, Calif) or normal rat IgG (Chemicon International Inc., Temecula, Calif) on days -4, -3, 3, and 10. The depletion of CD4 T cells and CD8 T cells were confirmed by flow-cytometry analysis (purity: >99%; data not shown).

In vitro T-cell differentiation

The *in vitro* differentiated T_H1/T_H2 and Tc1/Tc2 cells were prepared as described previously,¹⁸ restimulated, then subjected to an intracellular cytokine staining of IL-4 and IFN-γ¹⁹ or ELISA as previously described.¹⁸

Immunoprecipitation and immunoblotting

Immunoblotting for the detection of p50 and p65 nuclear factor-κB (NF-κB) subunits, GATA3, tubulin-α, nuclear factor of activated T cells (NFAT) 1, and NFAT2 was performed as described previously.^{18,20} The phosphorylation of inhibitory NF-κB (IκBα) was assessed by a phospho-IκBα detection kit (9100; New England

Biolabs, Ipswich, Mass). The detection of phosphorylation of mitogen-activated protein kinase (MAPK; extracellular signal-regulated kinase [ERK] 1 and ERK2) was performed as described.²¹

Electrophoretic mobility shift assay

Electrophoretic mobility-shift assays (EMSAs) were performed by using Gel shift Assay Systems (Promega, Madison, Wis) according to the methods previously described.²⁰

Quantitative real-time RT-PCR

Quantitative real-time RT-PCR was performed by using the ABI/PRISM 7900HT Sequence Detection System (Applied Biosystems, Foster City, Calif) and TaqMan Gene Expression systems (Applied Biosystems) as previously described.²⁰

Cell transfer of IκBαM-infected CD4 T cells

The generation of a retrovirus encoding a dominant-negative IκBα, IκBαM (pMX-IRES-IκBαM-hNGFR), and its infection into developing T_H2 cells was performed as described previously.²² The NC/Nga mice received anti-CD4 mAb on days -8 and -7 and were intraperitoneally injected with 50 μg antirat IgG_{2b} mouse IgG (G15-337; PharMingen) on days -4 and -2 to deplete the injected anti-CD4 mAb. The CD4 T cell-depleted mice were then injected intraperitoneally with 2 × 10⁷ T_H2 cells expressed IκBαM. The activation of NF-κB in the IκBαM-infected T_H2 cells was suppressed as previously described (data not shown).²²

RESULTS

T_H1/T_H2 and Tc1/Tc2 cell differentiation and cytokine production in NC/Nga mice

We first examined the ability of naive splenic T cells from NC/Nga mice maintained under SPF conditions to differentiate into T_H1/T_H2 and Tc1/Tc2 cells by cytoplasmic cytokine staining (Fig 1, A) and ELISA (Fig 1, B). BALB/c mice are known to be T_H2 prone mice, but no AD-like skin lesions normally develop under conventional conditions. The generation of T_H2 and Tc2 cells was significantly higher in NC/Nga mice than BALB/c mice (Fig 1, A). The number of T_H1 and Tc1 cells also slightly increased in the NC/Nga mice. The levels of IL-5 and IL-13 production in NC/Nga CD4 T_H2 cells and the levels of IFN-γ in NC/Nga T_H1 cells were significantly higher than those in BALB/c groups (Fig 1, B). No significant difference in the production of IL-4 was detected between NC/Nga and BALB/c CD4 T cells in the ELISA assay. In CD8 T cells, the production of IL-4, IL-5, and IL-13 was obviously higher in NC/Nga Tc2 cells. The production of IFN-γ also significantly increased in the Tc1 cells from NC/Nga mice. These results together suggest that the ability in NC/Nga mouse naive T cells to differentiate into T_H1/T_H2 and Tc1/Tc2 cells is higher than that in BALB/c naive T cells.

T-cell antigen receptor-induced calcium mobilization, ERK1/2 phosphorylation, and NF-κB activation in CD4 and CD8 T cells from NC/Nga mice

We next examined the T-cell antigen receptor (TCR)-mediated signaling pathways that play a crucial role in

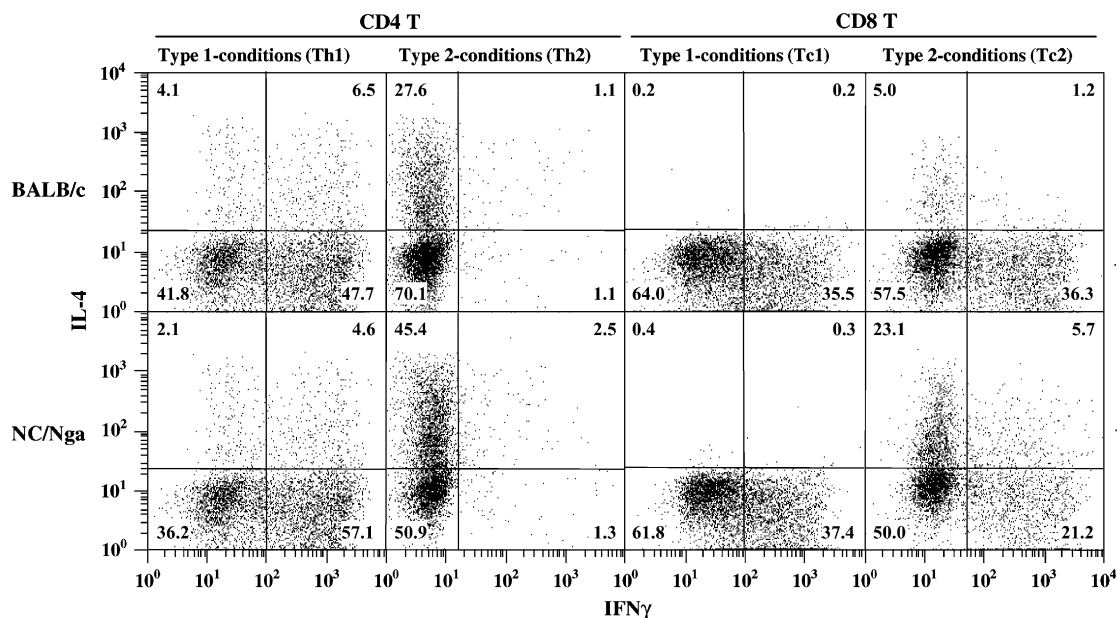
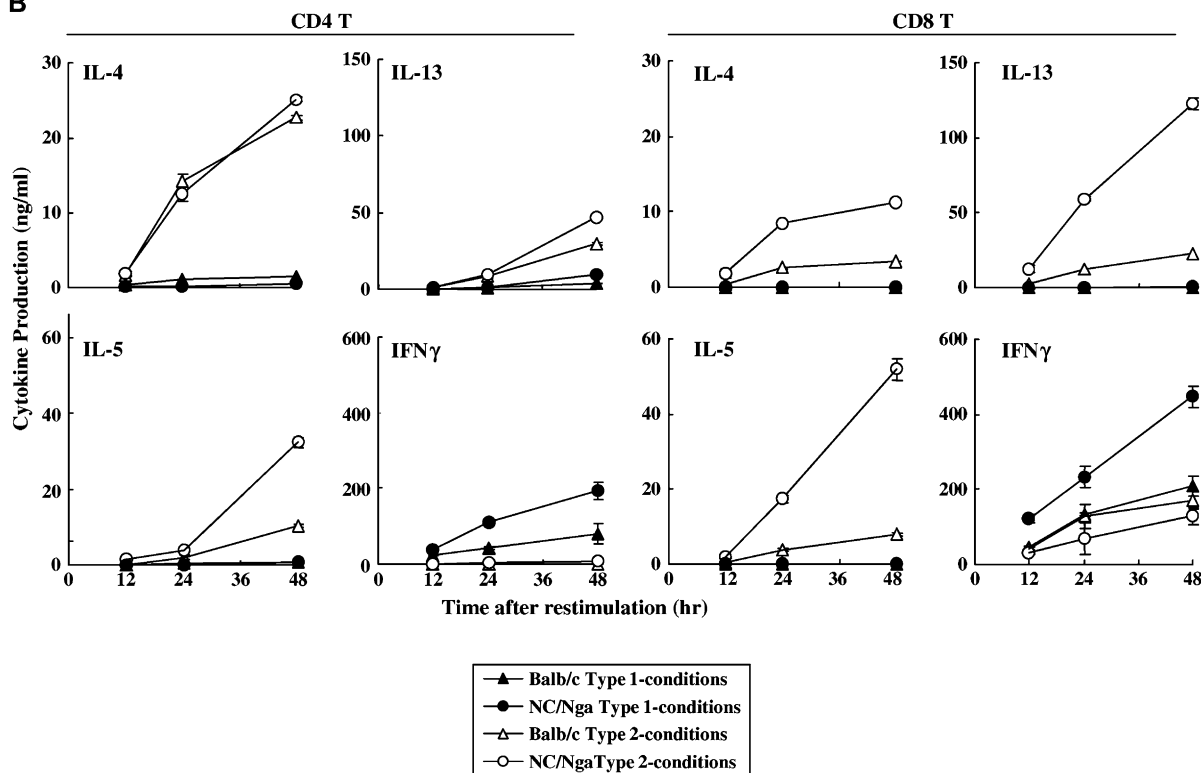
A**B**

FIG 1. Cytokine production profiles of *in vitro* differentiated T_H1/T_H2 cells and $Tc1/Tc2$ cells from BALB/c and NC/Nga mice. **A**, Representative IFN- γ /IL-4 profiles with the percentages of cells present in the each quadrant are shown. Five independent experiments were performed with similar results. **B**, The production of cytokines (IL-4, IL-5, IL-13, and IFN- γ) was determined by ELISA. Three independent experiments were performed with similar results.

T_H1/T_H2 cell differentiation and cytokine production. We studied the intracellular free-calcium ion concentration in CD4 T cells after TCR cross-linking, and comparable calcium mobilization was observed between NC/Nga and

BALB/c mice (see this article's Fig E1 in the Online Repository at www.jacionline.org). Next, T cells from NC/Nga and BALB/c mice were stimulated with immobilized anti-TCR antibody for several hours, and the

phosphorylation of ERK1/2, nuclear translocation of NF- κ B (p65 and p50), and phosphorylation of I κ B α were examined. The extent of the phosphorylation of ERK1 and ERK2 was significantly higher in both CD4 and CD8 T cells from NC/Nga mice than in BALB/c mice (Fig 2, A). The translocation of the p65 and p50 subunits of NF- κ B into the nucleus and the phosphorylation of I κ B α also increased in both CD4 and CD8 T cells from NC/Nga mice (Fig 2, B and C). It is interesting that I κ B α is phosphorylated in NC/Nga CD4 T cells even without any anti-TCR stimulation. Consequently, we more directly examined the activation of NF- κ B by an EMSA and found the NF- κ B DNA binding activity to be clearly higher in both CD4 and CD8 T cells from NC/Nga mice than in BALB/c mice, and the level of NF- κ B activation was enhanced after anti-TCR stimulation in either BALB/c or NC/Nga T cells (Fig 2, D). We performed a super-shift assay to confirm the specificity of the NF- κ B subunits. These results indicate that the levels of NF- κ B activation are higher in NC/Nga CD4 and CD8 T cells than in BALB/c T cells.

GATA3 and NFAT expression in developing T_H2 and Tc2 cells

We examined the induction of the expression of GATA3, NFAT1, and NFAT2 in developing T_H2 and Tc2 cells (see this article's Fig E2 in the Online Repository at www.jacionline.org). The results suggest that the expression of GATA3, NFAT1, and NFAT2 was not apparently different between NC/Nga and BALB/c mice, except for an increased GATA3 expression in NC/Nga Tc2 cells.

IL-2-induced and IL-4-induced STAT5 and STAT6 activation in CD4 and CD8 T cells from NC/Nga mice

Concurrently, the activation of cytokine-induced signaling pathways in developing T_H2 and Tc2 cells of NC/Nga mice was assessed (see this article's Fig E3 in the Online Repository at www.jacionline.org). No obvious difference in the levels and the time course of the phosphorylation of STAT5A, STAT5B, and STAT6 was detected between BALB/c and NC/Nga T cells, indicating that the capability of activation of IL-2-induced and IL-4-induced signaling pathways in developing T_H2 and Tc2 cells of NC/Nga mice is not enhanced.

Gene expression analysis between C57BL/6, BALB/c, and NC/Nga mice in T_H2 and Tc2 cells

To identify the genes that may be responsible for the hyperresponsive features in NC/Nga mouse T cells, we examined the differences in gene expression between BALB/c and NC/Nga mice using developing Tc2 cells and the GeneChip Mouse Genome array assay (Affymetrix, Santa Clara, Calif). We detected 54 known genes with transcription levels increased more than 4 times, and 25 known genes in which the levels decreased to less than 25% in NC/Nga mice in comparison with BALB/c mice (see this article's Fig E4 in the Online Repository at

www.jacionline.org). To quantify the difference more accurately, we assessed the mRNA expression of these genes in T_H2 and Tc2 cells from C57BL/6, BALB/c, and NC/Nga mice by quantitative real-time RT-PCR. As a result, the expression levels of 10 genes were found to selectively increase or decrease in NC/Nga mice in comparison with C57BL/6 or BALB/c mice. The quantification results are summarized in this article's Fig E5 in the Online Repository at www.jacionline.org). These results suggest that NC/Nga CD4 and CD8 T cells possess both an increased and decreased expression of a unique set of genes, although the pathophysiological consequences remain unclear.

Involvement of CD4 T cells in AD-like skin lesions induced by the topical injection of mite antigens in NC/Nga mice

Next, we induced dermatitis by repeated intradermal injections of mite antigens (Dp extract), a popular environmental antigen, into the ventral side of the ears. In this model, even under SPF conditions, severe dermatitis is reproducibly induced in NC/Nga mice.¹⁷ We observed a low-level increase in the ear thickness in BALB/c mice and significantly increased levels of ear thickness in NC/Nga mice (Fig 3, A). To determine the involvement of CD4 and CD8 T cells in the induction of AD-like skin lesions in NC/Nga mice, we administered anti-CD4 or anti-CD8 mAbs to deplete CD4 T or CD8 T cells before Dp extract injection. Normal rat IgG was used as a control. The depletion of CD4 T cells but not CD8 T cells significantly suppressed the ear thickness in NC/Nga mice (Fig 3, B). Histologic analyses with hematoxylin and eosin (HE) and toluidine blue staining indicated typical atopic dermatitis in the lesions of NC/Nga mice receiving Dp extract injection, and the severity of dermatitis was reduced in CD4 T-cell-depleted NC/Nga mice (see this article's Fig E6 in the Online Repository at www.jacionline.org). These results suggest that CD4 T cells are involved in the development of the AD-like skin lesions induced by the topical injection of Dp extract in NC/Nga mice.

Reduced NF- κ B activation in CD4 T cells results in the decreased AD-like skin lesions induced by the topical injection of mite antigens

To investigate the role of NF- κ B activation in CD4 T cells in the pathogenesis of AD-like skin lesions induced by the topical injection of Dp extract, NC/Nga T_H2 cells infected with a retrovirus encoding I κ B α M (a specific inhibiting molecule in NF- κ B activation^{22,23}) were prepared and transferred into CD4 T-cell-depleted NC/Nga mice, and the severity of the dermatitis was assessed. A significantly reduced ear thickness was observed in the mice receiving the I κ B α M-introduced T_H2 cells in comparison with those receiving mock-infected T_H2 cells (Fig 3, C). These results suggest that the levels of activation of NF- κ B in T_H2 cells (CD4 T cells) thus play a critical role in the pathogenesis of the dermatitis.

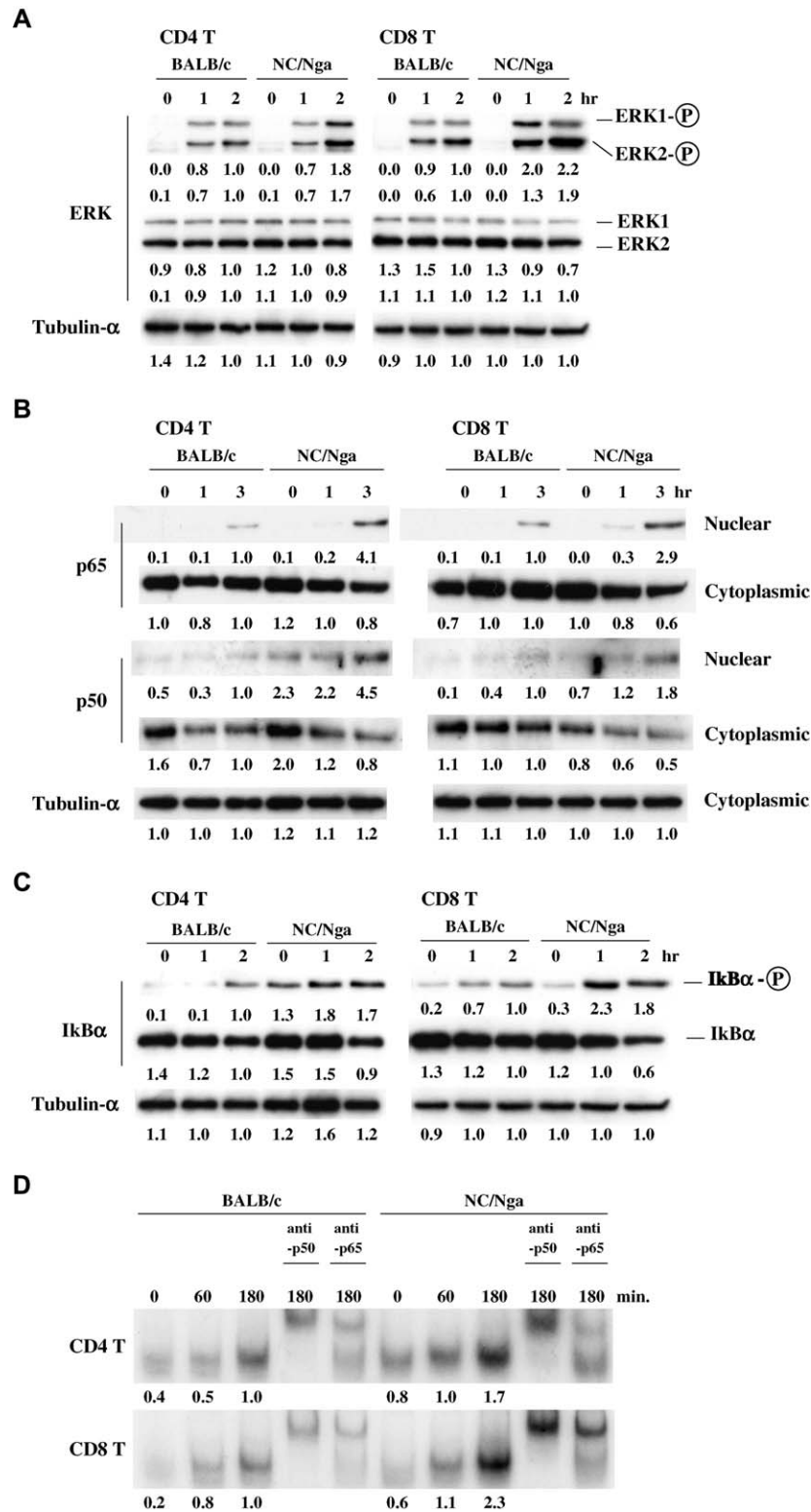


FIG 2. Increased activation of the ERK/MAPK cascade and the NF- κ B signaling pathway in CD4 T and CD8 T cells from NC/Nga mice. Freshly prepared splenic CD4 and CD8 T cells were stimulated with immobilized anti-TCR β mAb for the indicated times. Three independent experiments of each were performed with similar results. **A**, Immunoblotting using antiphospho-ERK, anti-ERK, and anti-tubulin- α antibodies. **B**, Immunoblotting of p65 and p50 subunit of NF- κ B. **C**, Immunoblotting of phospho-IkB α and IkB α . **D**, EMSAs with the NF- κ B probe. Stimulation of the cells was performed with immobilized anti-TCR mAb for the indicated times.

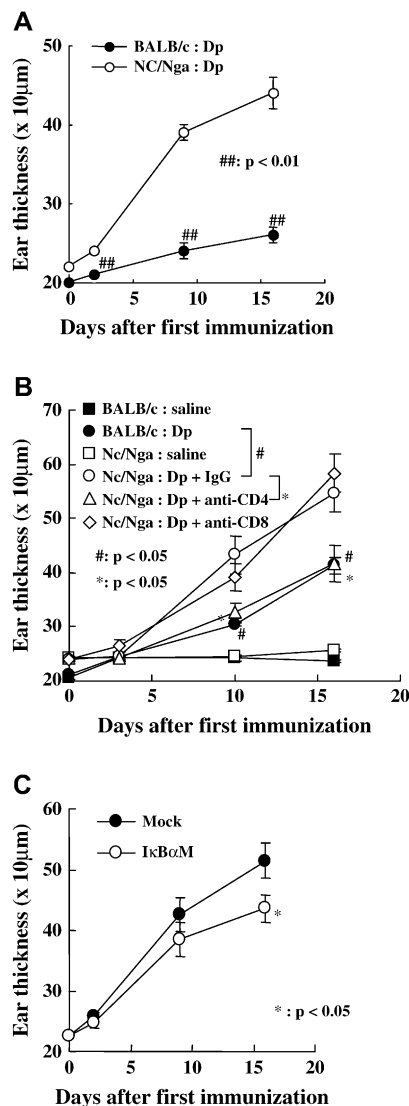


FIG 3. Involvement of CD4 T cells in the pathogenesis of AD-like skin lesions induced by the topical injection of mite antigens in NC/Nga mice. **A**, BALB/c and NC/Nga mice were injected with mite antigens (Dp extract) on days 0, 1, 7, 8, 14, and 15. Each symbol represents the mean $\pm$ SE of 5 mice. ## $P < .01$. **B**, BALB/c and NC/Nga mice were injected with saline or mite antigens (Dp extract) on days 0, 2, 8, 9, 14, and 15. Each symbol represents the mean $\pm$ SE of 5 mice. # $P < .05$, BALB/c Dp extract injection vs NC/Nga Dp extract injection; * $P < .05$, anti-CD4 injected group vs normal rat IgG-injected control group in NC/Nga mice with Dp extract injection. **C**, CD4 T-cell-depleted NC/Nga mice were injected with T_H2 cells infected with a mock or IkB α M-encoding retrovirus on day 0. Then the mice were injected with Dp extract as in A.

Increased T_H2 cytokine expression in draining lymph node T cells from NC/Nga mice with AD-like skin lesions induced by the topical injection of mite antigens

To investigate further the involvement of T_H2 cells in the AD-like skin lesions, the expression of T_H2 cytokines (IL-4, IL-5, and IL-13), a T_H1 cytokine IFN- γ , and

inflammatory cytokines (IL-6 and TNF- α) in T cells from the spleen, draining lymph nodes (DLs) and non-draining lymph nodes of BALB/c and NC/Nga mice were assessed by using quantitative real time RT-PCR (Fig 4, A). The mRNA expression of IL-4, IL-5, IL-13, and IL-6 in DL T cells increased in comparison with that in splenic T cells in either BALB/c or NC/Nga mice. The levels of IL-4, IL-5, IL-13, and IL-6 were significantly higher in NC/Nga mice. DL T-cell-specific decrease of IFN- γ and DL T-cell-specific increase of TNF- α were observed, but the differences were moderate. Concurrently, the expression levels of transcriptional factor GATA3, c-Maf, and T-box expressed T cells (T-bet) were assessed, and as expected, the selectively increased expression of GATA3 and c-Maf was detected in the DL T cells of NC/Nga mice (Fig 4, B). The increase was marginal in BALB/c mice. A robust expression of IL-4, IL-5, IL-13, and IL-6 was detected in DL CD4 T cells, but only a slight expression was seen in DL CD8 T cells. The levels were significantly higher in NC/Nga T cells. IFN- γ and TNF- α were expressed in both CD4 and CD8 T cells in either BALB/c or NC/Nga mice. Similarly, the preferential expression of GATA3 and c-Maf was observed in DL CD4 T cells but not in CD8 T cells (Fig 4, D). The activation level of NF- κ B and AP1 was found to increase in DL cells of NC/Nga mice (see this article's Fig E7 in the Online Repository at www.jacionline.org). Taken together, these results indicate that CD4⁺ T_H2 cells preferentially accumulated in the DL of NC/Nga mice with AD-like skin lesions.

DISCUSSION

The hyperresponsive feature of signaling in NC/Nga CD4 and CD8 T cells was selectively observed in the signal transduction pathways downstream of TCR but not of cytokine receptors, such as IL-2 receptor (IL-2R) and IL-4 receptor (IL-4R; see this article's Fig 2 and Fig E3 in the Online Repository at www.jacionline.org). Among the TCR-mediated signaling pathways, enhancement was observed in the Ras-MAPK cascade and NF- κ B signaling (Fig 2). It is well known that cytokines such as IL-12 and IL-4 play an important role in T_H1 and T_H2 cell differentiation, respectively.²⁴⁻²⁶ However, the activation of TCR-mediated signaling pathways is also indispensable for both T_H1 and T_H2 cell development.²⁷ In particular, the extent of the TCR-mediated activation of the Ras/MAPK cascade controls T_H2 cell differentiation and ovalbumin-induced allergic airway inflammation.^{19,28} These previous findings are consistent with the observation in this report that NC/Nga T cells show an enhanced activation of the Ras/MAPK cascade (Fig 2, A), and the levels of T_H2 cell differentiation and the production of T_H2 cytokines were enhanced in T_H2 and Tc2 cells from NC/Nga mice (Fig 1). Moreover, the activity of AP1, a downstream target of the Ras/MAPK cascade,²⁹ appeared to increase selectively in draining lymph node T cells in NC/Nga mice after the topical injection of mite antigens (see this

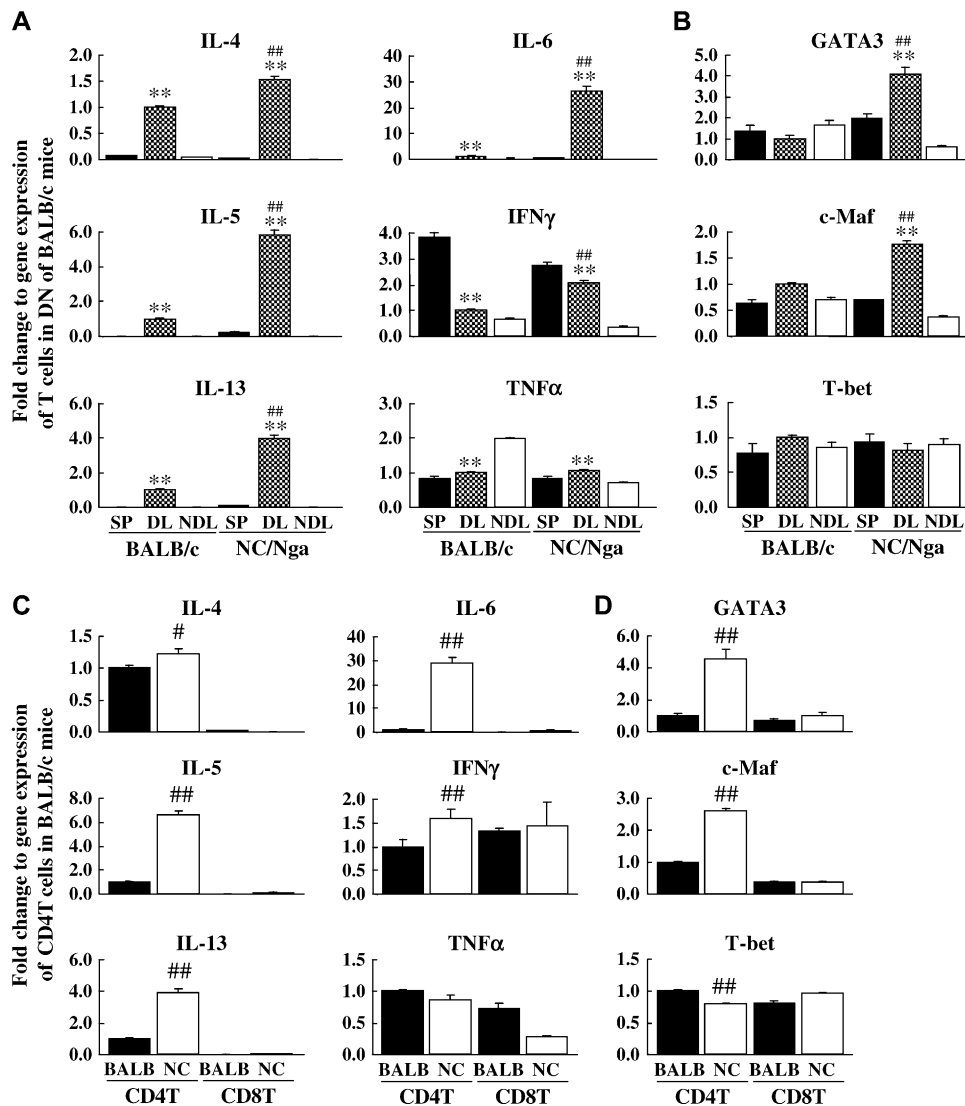


FIG 4. Increased mRNA expression of the T_H2 cytokine genes, GATA3 and c-Maf, in DL CD4 T cells from NC/Nga mice with AD-like skin lesions induced by topical application of mite antigens. **A** and **B**, T cells were prepared from the spleen (SP), DL (submandibular lymph node), and nondraining lymph node (NDL, mesenteric lymph node) of Dp extract-injected mice on day 16. The mRNA expression was analyzed by quantitative real-time PCR. The expression was presented as the fold change to that in the spleen T cells from Dp extract-injected BALB/c mice. Two independent experiments were performed with similar results. ** $P < .01$, DL vs spleen in BALB/c or NC/Nga; ## $P < .01$, NC/Nga vs BALB/c. **C** and **D**, CD4T and CD8T cells were subjected to quantitative real-time PCR. # $P < .05$, NC/Nga vs BALB/c; ## $P < .01$, NC/Nga vs BALB/c.

article's Fig E7 in the Online Repository at www.jacionline.org). In T_H2 cells, the activation of the Ras-ERK MAPK cascade is induced by stimulation of TCR but not induced by stimulation with IL-2 or IL-4 alone.¹⁹ Therefore, the intrinsic defects in T cells of NC/Nga mice appear to exist selectively in the TCR-mediated signal transduction pathways.

It is well established that NF- κ B plays a crucial role in the innate and acquired immune responses and chronic inflammation.³⁰⁻³³ The NF- κ B activity is crucial for the development of T_H1 responses.³⁴⁻³⁶ NF- κ B is also known to be a critical transcription factor that regulates

T_H2 cell differentiation and T_H2 -dependent airway inflammation.^{20,37-39} We detected an increased activation of the NF- κ B signaling pathway in normal splenic CD4 and CD8 T cells from SPF NC/Nga mice without AD-like skin lesions (Fig 2, B-D). Interestingly, an increased phosphorylation of I κ B α and an increased NF- κ B binding activity were both detected without anti-TCR stimulation. The increased NF- κ B activity was detected in the draining lymph node T cells from NC/Nga mice receiving Dp injection (see this article's Fig E7 in the Online Repository at www.jacionline.org). Thus, it is likely that the AD-like skin lesions and the increased T_H2 cytokine expression

in the draining lymph node T cells would be at least in part a consequence of the enhanced activation of the NF- κ B signaling pathway in NC/Nga T cells.

In the current study, we demonstrate that T_H2 cells play a crucial role for the pathogenesis of AD-like skin lesions induced by mite antigens in NC/Nga mice (Figs 3 and 4). This appears to be contradictory to the results in a report using STAT6-deficient NC/Nga mice.¹⁶ This group investigated the AD-like dermatitis induced in NC/Nga mice maintained under conventional conditions. This AD-like dermatitis could be induced by many different unknown environmental factors, and various inflammatory cell types, including mast cells, eosinophils, and basophils as well as T_H2 cells are known to be involved. This may be the reason why Yagi et al¹⁶ did not find a critical role of T_H2 cells in the spontaneously induced dermatitis in NC/Nga mice.

We detected an increased expression of vitamin D receptor and NACHT leucine rich repeat and PYD containing 6 (Nalp6) in NC/Nga T cells in comparison with either C57BL/6 or BALB/c T cells (see this article's Fig E5 in the Online Repository at www.jacionline.org). 1 α , 25-Dihydroxyvitamin D3 has been reported to act directly on naive CD4 T cells, thereby enhancing the development of T_H2 cells.⁴⁰ Nalp6 synergistically activates NF- κ B with the expression of a caspase activating recruitment domain (CARD)-containing molecule apoptosis-associated speck-like protein containing a caspase recruitment domain (ASC).⁴¹ Thus, although no direct evidence exists, the increased expression of vitamin D receptor and Nalp6 in NC/Nga T cells may explain the increased T_H2 cell differentiation and NF- κ B hyperactivation in NC/Nga T cells.

From a therapeutic point of view, NF- κ B has been considered to be a good target molecule for chronic inflammation.⁴²⁻⁴⁴ Our results on the pathogenesis of AD-like skin lesions in NC/Nga mice (Fig 3, C) indicate that the inhibition of the NF- κ B activity may also be effective in the treatment of acute phase of AD. RelB-deficient mice develop T-cell-dependent skin lesions similar to those in human AD.⁴⁵ In addition, the I κ B α deficiency results in widespread dermatitis accompanied with sustained NF- κ B responses.⁴⁶ The administration of NF- κ B decoy improved partially the spontaneously induced AD-like lesions in NC/Nga mice.⁴⁷

There are various possible strategies to develop selective inhibitors for NF- κ B, including (1) inhibiting NF- κ B DNA-binding by small molecules or nonfunctional oligonucleotides, (2) inhibiting nuclear translocation, (3) stabilizing I κ B α by regulating its phosphorylation and ubiquitin-dependent degradation, and (4) targeting upstream kinases such as I κ B kinase (IKK). Although the inhibition of the NF- κ B activity may induce some side effects including hepatotoxicity and opportunistic infections, the identification of a key component for specific inflammatory diseases and the development of specialized method of the treatment would help to solve these problems. In particular, therapeutic ointment can be used for patients with AD, and very limited systemic side effects are observed.

We thank Ms Kaoru Sugaya, Hikari Asou, and Satoko Norikane for their excellent technical assistance.

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