

Characterization of Spontaneous Dermatitis in Beige Rats with IgE Hyperproduction

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Key Words

Atopic dermatitis animal model · Fur mites · Helper T cells, type 1 · Helper T cells, type 2

Abstract

Background: Atopic dermatitis (AD) is a common skin disease characterized by chronic recurrent eczematous lesions, but its exact etiology and mechanism are unclear. We found that beige rats (*DAbg/bg*), a mutant model of Chediak-Higashi syndrome, develop skin lesions characterized by pruritus, excoriation, erosion and alopecia. We describe the beige rat and examine its possible usefulness as an AD model. **Methods:** Beige rats of 4, 8, 13, 16, 26 and 52 weeks were used. Histological analysis of the skin was performed. Plasma IgE and cytokines were measured. Th1 and Th2 cytokines and RANTES mRNA expression of skin and lymph nodes were evaluated. Passive cutaneous anaphylaxis (PCA) reactions were examined, and maximization tests were conducted. **Results:** Skin lesions begin to develop with increases in serum IgE levels and the expression of IL-4 mRNA in the lymph node and skin. Histologically, skin lesions are characterized by acanthosis, ulceration and inflammatory cell infiltration in the dermis. Inflammatory cells con-

sist of CD3+, CD4+, ED1+, ED2+ and I-A+ mononuclear cells, eosinophils, degranulated mast cells and neutrophils accompanying interleukin (IL)-4, interferon (IFN)- γ and RANTES mRNA expressions of the skin. Inflammatory cells are reduced during chronification with decreased expressions of IL-4, IFN- γ and RANTES mRNA. In addition, the rats show a high sensitivity to PCA reactions and maximization tests. **Conclusions:** Our results show that some of the skin lesions of beige rats are morphologically similar to human AD, being characterized by inflammatory cell composition in the acute phase, and increased IgE and RANTES levels. However, the inflammatory process and cytokine expression pattern are different from those in human AD.

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Introduction

Atopic dermatitis (AD) is a common skin disease characterized by chronic recurrent eczematous lesions [1, 2]. Many patients have an anamnesis and a familial history of some allergic diseases. The pathogenesis of AD is thought to be involved with immunological abnormalities [1, 2].

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Histopathologically, skin lesions are characterized by the infiltration of mononuclear cells with sparse eosinophils [3]. Such cells consist of macrophages and activated helper T cells bearing CD3, CD4 and MHC class II [4]. Helper T cells are divided into type 1 (Th1) and type 2 (Th2) by their cytokine secretion pattern [2]. Th1 cells secrete interleukin (IL)-2 and interferon (IFN)- γ that play an important role in cell-mediated immunity. Th2 cells secrete IL-4 and IL-5 that induce humoral immunity [2]. In the dermis of acute AD lesions, the cytokine milieu is much more Th2 skewed. CD4+ activated T cells predominantly infiltrate the dermis. However, the chronic phase of AD is reflected by a Th1 cytokine milieu [5–7]. In addition, dendritic cells and macrophages expressing Fc ϵ RI are increased in the epidermis and dermis with eosinophils and T cells [5, 8].

In acute lesions of AD, many eosinophils infiltrate the upper dermis. This abnormal migration and accumulation of eosinophils create inflammation and contribute to tissue damage [9]. RANTES, which is a CC chemokine with chemotactic and activated activity of eosinophils, is secreted by lymphocytes and monocytes [10]. RANTES mRNA and protein have been detected in rashes and non-ruptive lesions of AD [11, 12].

AD is characterized by allergen-specific IgE production with many patients showing an increase in general IgE levels, suggesting the involvement of an IgE-mediated type I allergy [1]. Mast cells are the central cells of type I allergy induced by the release of primary and secondary mediators. Degranulated mast cells have been reported to be prominent in acute dermal lesions of AD [13]. AD was thought to be a manifestation of type I allergy, but it is known that type IV allergy also plays an important role [14]. Recently, an important role in the induction is suggested for IgE-bearing epidermal Langerhans and non-IgE-bearing mast cells in human AD [5]. However, the precise pathogenesis of AD has not been completely clarified. A suitable animal model is needed to elucidate its pathogenesis. There are recent reports of NC/Nga and DS-Nh mice that suffered from AD-like dermatitis with IgE hyperproduction, but there are still few studies of an animal model [15–18]. Beige rats (*DAbg/bg*) are a suitable animal model of Chediak-Higashi syndrome [19, 20]. We found that almost all rats of this strain incidentally exhibit pruritus, hemorrhage, excoriation, erosion, crusts and alopecia in the skin [20]. Inflammatory skin lesions also develop with age. In this study, we report the characteristics of the skin lesions in the beige rat and its possible usefulness as an animal model of AD.

Materials and Methods

Animals

Beige rats (*DAbg/bg*) are descended from a light-colored mutant male rat found among DA rats (*DA+/+*) that originated from the Australian National Institute of Genetics as an inbred strain. This mutant line has been maintained by brother and sister mating under conventional housing conditions at the Research Institute of Drug Safety, Setsunan University. The rats in the present study were housed in a conventional air-conditioned animal room, fed a standard laboratory diet and supplied with water ad libitum.

We studied each group of rats as follows:

(1) Microscopic examination. Subjects consisted of beige rats aged 4, 8, 13, 16, 26 and 52 weeks (25 males, 25 females) and age-matched DA rats (25 males, 25 females). Their skin surface was also examined to detect ectoparasites using a surgical microscope (Operation Microscope 700, Konan Medical Inc., Nishinomiya, Japan).

(2) ELISA. Beige rats of 4, 8, 13, 16, 26 and 52 weeks of age (30 males).

(3) RT-PCR. Beige rats of 4, 8 and 26 weeks of age (15 males).

(4) Passive cutaneous anaphylaxis (PCA) reaction. Beige rats of 13 weeks of age (5 males) and age-matched DA rats (5 males). In addition, 10 male Sprague-Dawley [SD; Crj: CD(SD), Charles River Japan, Shiga, Japan] rats were used.

(5) Maximization test. Beige rats of 13 weeks of age (10 males) and age-matched DA rats (10 males).

The Committee for Ethical Use for Experimental Animals at Setsunan University approved all studies.

Light Microscopy

Rats were given gross examinations, anesthetized with ether and killed by exsanguination from the abdominal aorta. Skins were stretched on a flat board and fixed in 10% phosphate-buffered formalin (pH 7.4). Tissue samples of the skin were trimmed, dehydrated in a graded series of ethanol, embedded in paraffin, and 4- μ m-thick sections were stained with hematoxylin and eosin. Representative sections were also stained with toluidine blue for metachromasia of mast cell granules and by the luna method for eosinophil granules. For immunohistochemistry, skins were stretched on a flat board, fixed in IHC zinc fixative (BD Bioscience, San Jose, Calif., USA), dehydrated in a graded series of ethanol, embedded in paraffin and cut into 4- μ m-thick sections. Antihuman CD3 (polyclonal antibody; Dako, Glostrup, Denmark), antirat CD4 (monoclonal antibody W3/25; Serotec Ltd., Oxford, UK), antirat CD8 (monoclonal antibody OX-8; Serotec Ltd.), antirat ED1 (monoclonal antibody ED1; Serotec Ltd.), antirat macrophage (monoclonal antibody ED2; Serotec Ltd.) and anti-I-A (monoclonal antibody OX-6; Serotec Ltd.) were used for primary antibodies. All tissue sections were deparaffinized and dehydrated, rinsed in Tris-buffered saline with Tween 20 (TBST, pH 7.6), treated with 1% hydrogen peroxide and rinsed in TBST. Slides were incubated in bovine serum albumin for 5 min, and primary antibodies overnight at 4°C. Next day, slides were rinsed in TBST and treated with Simple Stain Rat MAX PO (Multi) (Nichirei, Tokyo, Japan) for 30 min at room temperature. Slides were rinsed using TBST, and diaminobenzidine solution containing 0.01% hydrogen peroxide was used for the peroxidase coloring reaction. The slides were then counterstained with Mayer's hematoxylin.

Enzyme-Linked Immunosorbent Assay

Rats were bled from the aorta, and serum was collected. Total IgE levels were measured by a sandwich ELISA using a Morinaga rat IgE kit (Morinaga Co. Ltd., Yokohama, Japan). The sensitivity for the assay was 1 ng/ml. IL-2, IL-4 and IFN- γ levels were measured by a sandwich ELISA using Wako IL-2, IL-4 and IFN- γ ELISA kits (Wako Pure Chemical Industries Ltd., Osaka, Japan). The sensitivities for the assay were 10, 15 and 13 pg/ml.

RNA Preparation

Skin samples (intact skin from 4-week-old rats, erupted skin from 8-week-old rats, alopecia skin without eruption from 30-week-old rats) and submandibular lymph nodes were collected. Messenger RNA was immediately isolated from the tissue by the guanidinium thiocyanate method using a Quick Prep Micro mRNA Purification Kit (Amersham Biosciences Corp., Piscataway, N.J., USA) with a Polytron PT1200CL (Kinematica AG, Littau, Switzerland).

Reverse-Transcription Polymerase Chain Reaction

A cDNA was synthesized from mRNA using a First-Strand cDNA Synthesis Kit (Amersham Biosciences Corp.). The specific primers for rat GAPDH, IL-2, IL-4, IFN- γ , and RANTES were used. Primer sequences were as follows:

IL-2: 5' primer, 5' ATGTACAGCATGCAGCTCGCATCC,
3' primer, 5' TTACTGAGTCATTGTTGAGATGAT;
IL-4: 5' primer, 5' TGATGGGTCTCAGCCCCACCTTGC,
3' primer, 5' CTTTCAGTGTGTGAGCGTGACTC;
RANTES: 5' primer, 5' GAAGATCTCTGCAGCTGCATCCCT,
3' primer, 5' CTTGAACCACTTCTTCTCTGGG;
IFN- γ : 5' primer, 5' ACACGCCGCGTCTTGGTTTT,
3' primer, 5' CGCTTCCTTAGGCTAGATTCTGG;
GAPDH: 5' primer, 5' ACCACAGTCCATGCCATCAC,
3' primer, 5' TCCACCACCCTGTTGCTGTA.

GAPDH, IL-2 and RANTES were used for one-step PCR. IL-4 and IFN- γ determination was conducted using two-step PCR. The PCR reaction mixture for GAPDH, IL-2, RANTES and the first-step PCR of IL-4 and IFN- γ consisted of 1 μ g of cDNA, 5' and 3' primers (50 mM each primer), 0.2 mM dNTP mixture, 10 mM Tris-HCl (pH 8.3), 50 mM KCl, 5 mM MgCl₂ and 1.25 units *Thermus aquaticus* DNA polymerase (Takara Co. Ltd., Shiga, Japan). The samples were subjected to 26 repeated cycles after an initial denaturing step (4 min at 94°C), each cycle consisting of 3 stages: denaturation for 1 min at 94°C, annealing for 1 min at 60°C and extension for 1 min at 72°C. Second-step PCRs for IL-4 and IFN- γ were used with 0.25 μ l of the first PCR product as a template, and a total 50- μ l volume of the same PCR reaction mixture and primers. The samples were subjected to 16 repeated cycles for IL-4 and 14 for IFN- γ . The amplified DNA products were analyzed by electrophoresis in a 2.0% agarose gel stained with ethidium bromide and viewed under UV transillumination.

PCA Reaction Test

Each of 5 DA+/+ and beige rats was injected subcutaneously with 1 ml aluminum hydroxide gel containing 2 mg ovalbumin and intraperitoneally with a 0.5-ml subcutaneous injection of pertussis inactive microorganism suspension (2×10^{10} /ml). Three weeks later, the rats were bled from the aorta, and serum was collected. SD rats [Crj: CD(SD)] were used as recipients. According to the method of Mota and Wong [21], the animals were sensitized with intradermal injections of 0.1 ml antiserum dilution ($\times 2$, $\times 4$, $\times 8$ and $\times 16$) at differ-

ent sites on each side of the dorsal skin. They were challenged 48 h later by intravenous injections of 1 ml saline containing 1 mg ovalbumin and 1% Evans blue solution. Thirty minutes later, the animals were killed by exsanguination from the abdominal aorta under deep anesthesia. The pigmented area was measured from the inner side of the skin. An area more than 5 mm in diameter was regarded as a positive reaction.

Maximization Test

The experiments were conducted according to the maximization test method described by Magnusson [22] and Magnusson and Kligman [23]. Induction was performed in two different ways; i.e. first by intradermal injection and secondly, 1 week later, by topical application over the injection sites. For the intradermal injections, an area of about 24 cm² of dorsal skin in the scapular region was shaved. A series of 2 intradermal injections (0.05 ml each) were made simultaneously in the shaved area. The following preparations were injected: 2 injections with Freund's complete adjuvant and saline (1:1), 2 injections with 0.1% dinitrochlorobenzene (DNCB) with corn oil and 2 injections with 0.1% DNCB/corn oil and Freund's complete adjuvant/saline (1:1). Six days after the intradermal injections, the dorsal skin in the scapular regions was shaved again. On the following day, the induction by topical application was performed with 0.5 g of 10% SDS in white petrolatum. The next day, an approximately 2×4 cm patch of lint cloth was loaded with 0.2 g of 0.1% DNCB in white petrolatum. The loaded patch was placed over the sites of the intradermal injections. The patch was covered with a surgical bandage and sponge pad, and secured in place with an elastic bandage wrapped around the animal for 48 h. After 14 days, the topical challenge was carried out on the animal's flank, which was clipped free of hair. Lint cloths (2×2 cm) were loaded with 0.1 ml of 0.625, 0.125, 0.25 and 0.5% DNCB in white petrolatum. The patches were covered with surgical bandages and sponge pads, and secured in place with an elastic bandage wrapped around the animal for 24 h. Skin reactions after the challenge were evaluated at 0 and 24 h following removal of the patches. The following ordinal grading scale was used: 0 = no visible change, 1 = discrete or patchy (slight) erythema, 2 = moderate and confluent erythema, and 3 = intense (severe) erythema and swelling [23].

Results

Gross Findings

All beige rats over 8 weeks old developed pruritus, hemorrhage, excoriation, erosion, crusts and alopecia on their skin. The earliest skin lesions were detected as single or multiple small hemorrhages and erosions with alopecia around the neck. The lesions extended caudally to involve the skin of the postscapular dorsum with multiple ulcers caused by scratching [20]. A surgical microscopic examination revealed a few fur mites (*Radfordia ensifera*) on the skin surface.

Microscopic Findings

Acute lesions were initially observed in 8-week-old rats. With aging, the acute and chronic lesions became

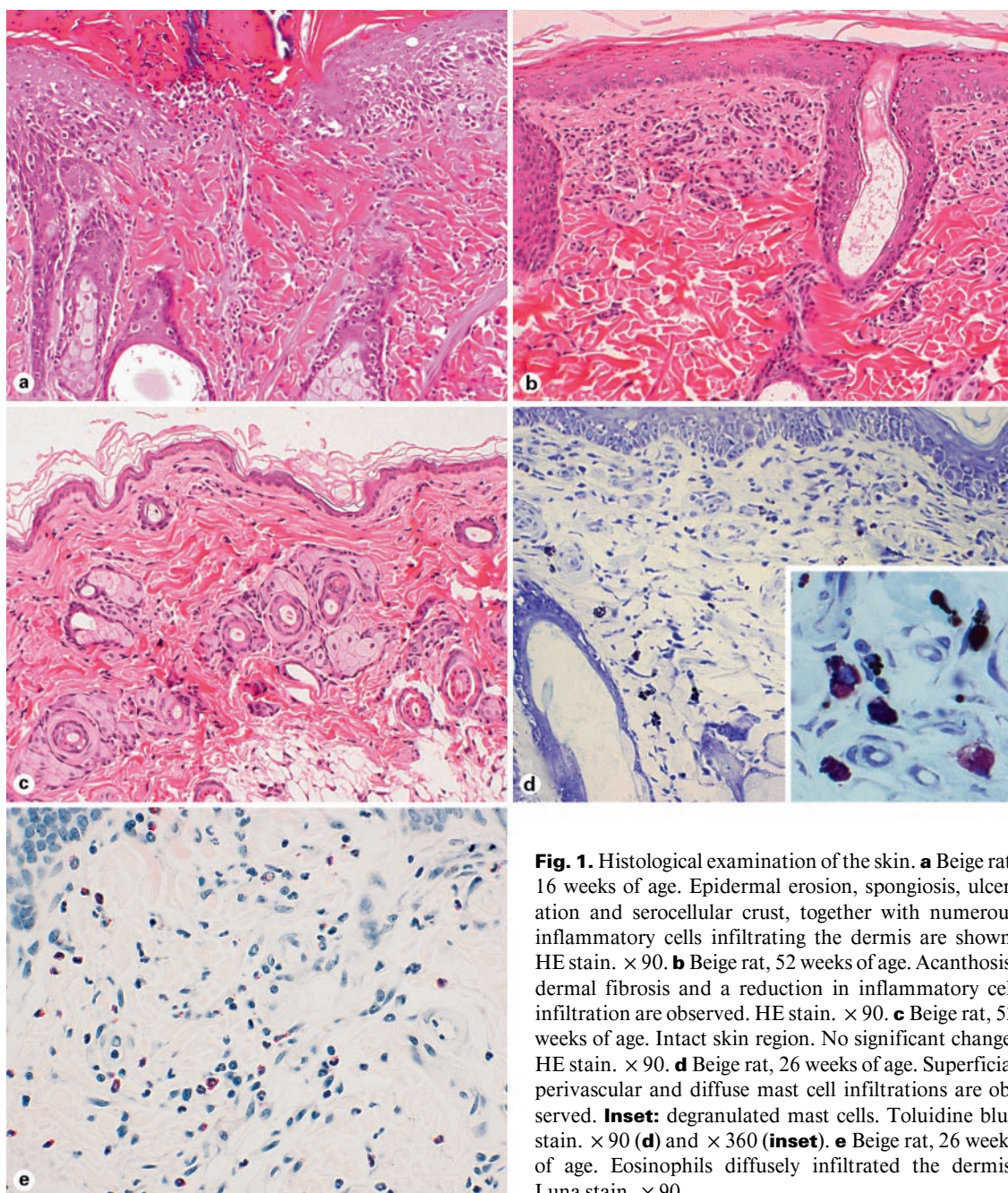


Fig. 1. Histological examination of the skin. **a** Beige rat, 16 weeks of age. Epidermal erosion, spongiosis, ulceration and serocellular crust, together with numerous inflammatory cells infiltrating the dermis are shown. HE stain. $\times 90$. **b** Beige rat, 52 weeks of age. Acanthosis, dermal fibrosis and a reduction in inflammatory cell infiltration are observed. HE stain. $\times 90$. **c** Beige rat, 52 weeks of age. Intact skin region. No significant change. HE stain. $\times 90$. **d** Beige rat, 26 weeks of age. Superficial perivascular and diffuse mast cell infiltrations are observed. **Inset:** degranulated mast cells. Toluidine blue stain. $\times 90$ (**d**) and $\times 360$ (**inset**). **e** Beige rat, 26 weeks of age. Eosinophils diffusely infiltrated the dermis. Luna stain. $\times 90$.

intermingled, causing the expansion of the region with these lesions. In acute lesions that showed epidermal erosion, ulceration and serocellular crust, many inflammatory cells had infiltrated the dermis as compared with the intact skin of beige rats (fig. 1a, c). A number of neutrophils and eosinophils infiltrated just beneath the epidermal layer with ulceration and erosion. In the area apart

from the ulcerative lesion, dermal lesions were seen that included superficial perivascular and diffuse inflammatory cell infiltrations of mast cells (fig. 1d), macrophages, lymphocytes and eosinophils (fig. 1e). Many of the mast cells were degranulated (fig. 1d). Immunohistochemically, many ED1+, ED2+, I-A+ and CD3+ cells had diffusely infiltrated the upper dermis (fig. 2, 3). I-A+ and CD3+

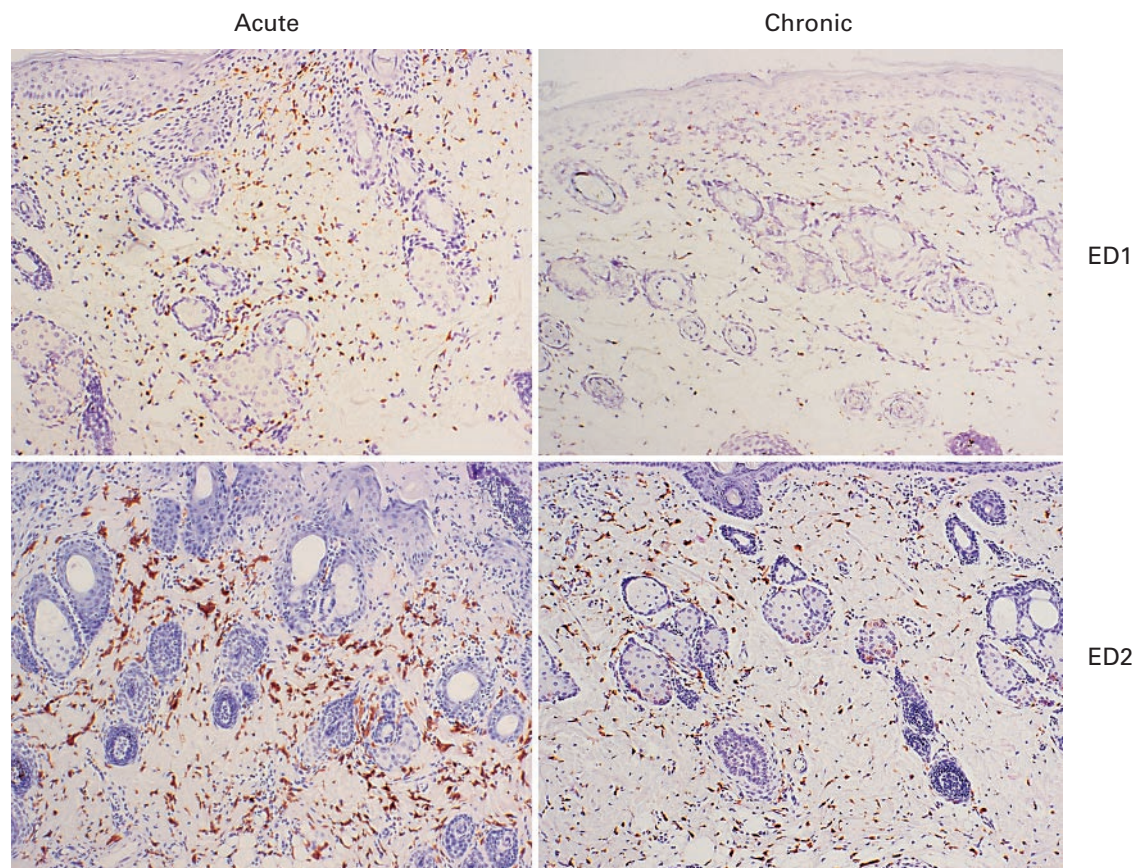


Fig. 2. Immunohistochemical stain of the skin. Beige rat, 26 weeks of age. Many ED1+ and ED2+ cells diffusely infiltrate the upper dermis in the acute lesions, but they are fewer in number in the chronic lesions. Mayer's hematoxylin counterstain. $\times 180$.

cells were also frequently observed within the epidermis (fig. 3). CD4+ cells often infiltrated the upper dermis, but single CD8+ cells were also occasionally seen there (fig. 3).

Chronic lesions were characterized by moderate acanthosis, orthokeratotic hyperkeratosis, dermal fibrosis and a lower level of inflammatory cell infiltration localized in the superficial dermis as compared with the intact skin of beige rats (fig. 1b, c). The number of mast cells was unchanged compared with acute lesions. Many mast cells were intact, but some were degranulated. Immunohistochemically, the number of I-A+, ED1+, ED2+, CD3+ and CD4+ cells decreased, but that of CD8+ cells was little changed (fig. 2, 3).

Reverse-Transcription Polymerase Chain Reaction

In the skin, IL-4 and IFN- γ mRNA were detected in acute lesions, but were undetectable in the areas of chronic lesions or intact skin (fig. 4). RANTES mRNA was detected from intact skin to chronic lesions, but its expression decreased toward the chronic lesions (fig. 4). IL-2 expression was undetectable in the skin at any stage of lesions.

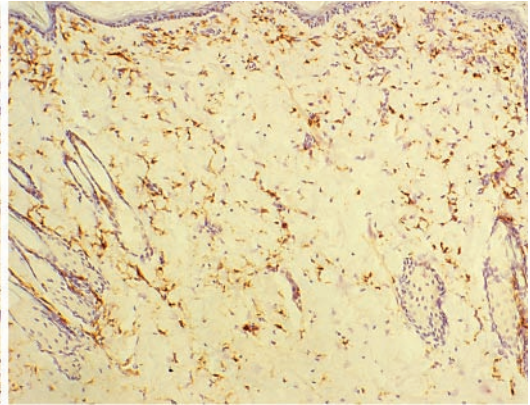
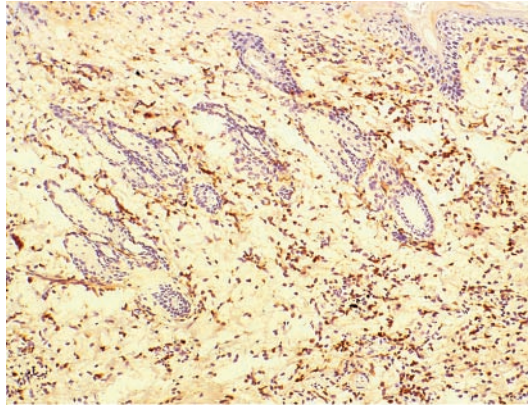
In the lymph nodes, IL-4 mRNA was detected from the acute to the chronic lesions (fig. 4). IFN- γ and RANTES mRNA were detected from intact skin to the chronic lesions (fig. 4). IL-2 expression was undetectable in the skin at any stage of lesions.

Plasma Total IgE, IL-2, IL-4 and IFN- γ

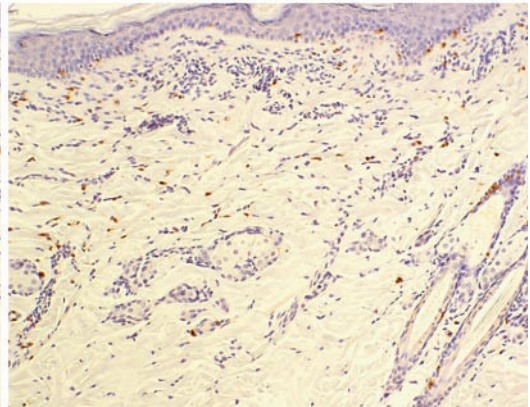
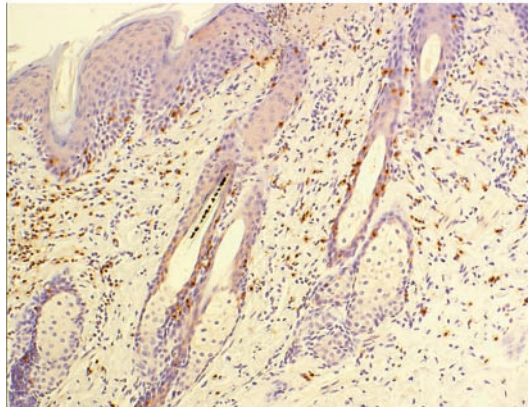
The plasma levels of total IgE were low at 4 and 8 weeks (fig. 5), had slightly increased at 13 weeks and were

Acute

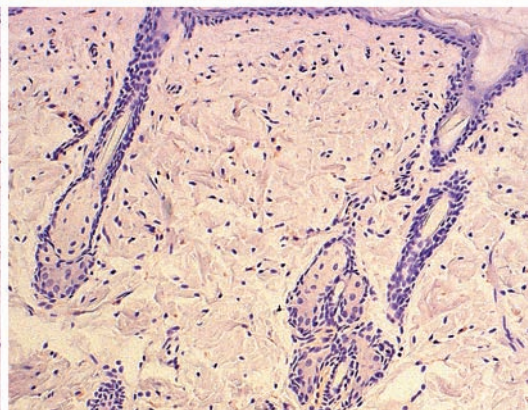
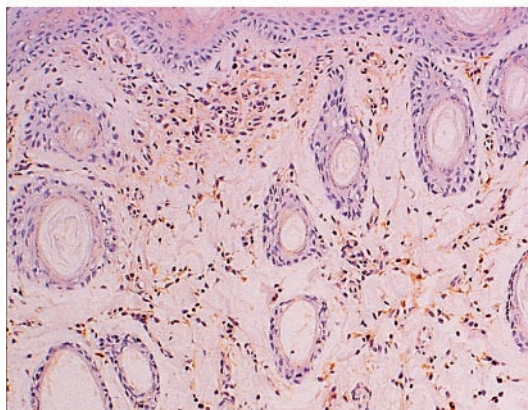
Chronic



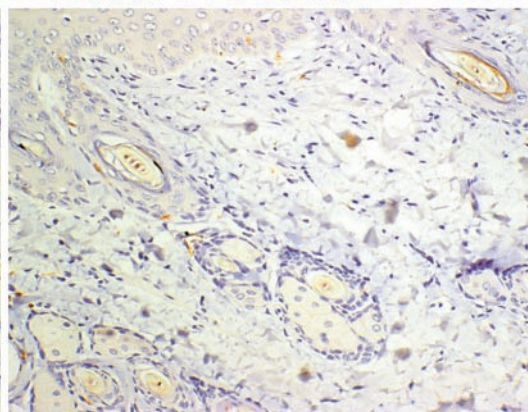
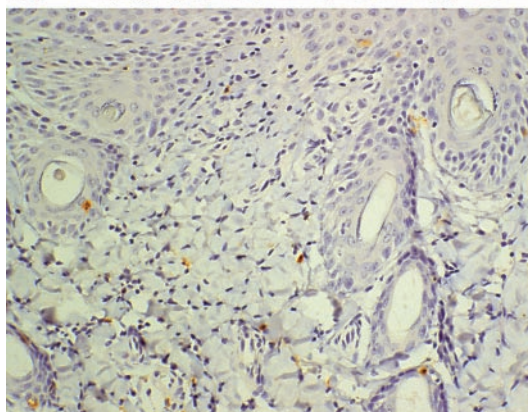
I-A



CD3



CD4



CD8

3

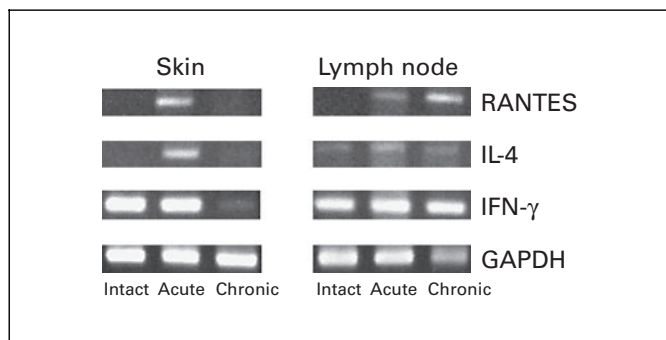


Fig. 4. RT-PCR analysis of RANTES, IL-4, IFN- γ and GAPDH in the lymph nodes and skin of beige rats. IL-4 and IFN- γ mRNA are detected in the acute skin lesions but are undetectable in the chronic lesions and intact skin. RANTES mRNA is detected from the intact skin to chronic lesions, but its expression declines in the chronic phase. In the lymph nodes, IL-4 mRNA is detected in lesions from the acute to chronic stages. IFN- γ and RANTES mRNA are detected from the intact to chronic stages.

markedly elevated from 16 weeks of age, corresponding to the advance in the severity of the skin lesions. The elevated level persisted until 52 weeks. However, plasma IL-2, IL-4 and IFN- γ levels did not change significantly from the intact skin to the chronic lesions (data not shown).

PCA Reaction and Maximization Test

The PCA response was remarkably induced in beige rats, although even ovalbumin induced a slight PCA response in DA+/+ rats. The delayed hypersensitivity response of beige rats using the maximization test was higher than that of DA rats. In beige rats, 0.5% DNCB showed a positive response in the form of moderate to strong erythema (score 2–3), and 0.25% DNCB induced a slight to moderate positive response (fig. 6). However, in DA+/+ rats, only 0.5% DNCB provoked a slight to moderate positive response (score 1–2; fig. 6).

Fig. 3. Immunohistochemical stain of the skin. Beige rat, 26 weeks of age. A number of I-A+ and CD3+ cells infiltrate the upper dermis in the acute lesions, but these cells decrease in number in the chronic lesions. In the acute lesions, CD4+ cells are frequently present in the upper dermis, but single CD8+ cells are also occasionally seen in the chronic lesions. CD4+ cells decrease, whereas CD8+ cells show little change. Mayer's hematoxylin counterstain. $\times 180$.

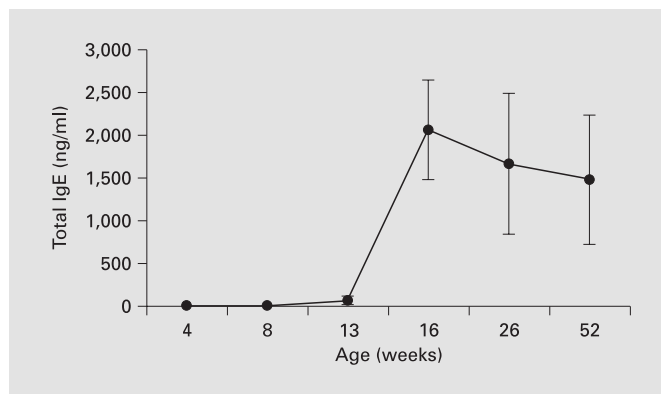


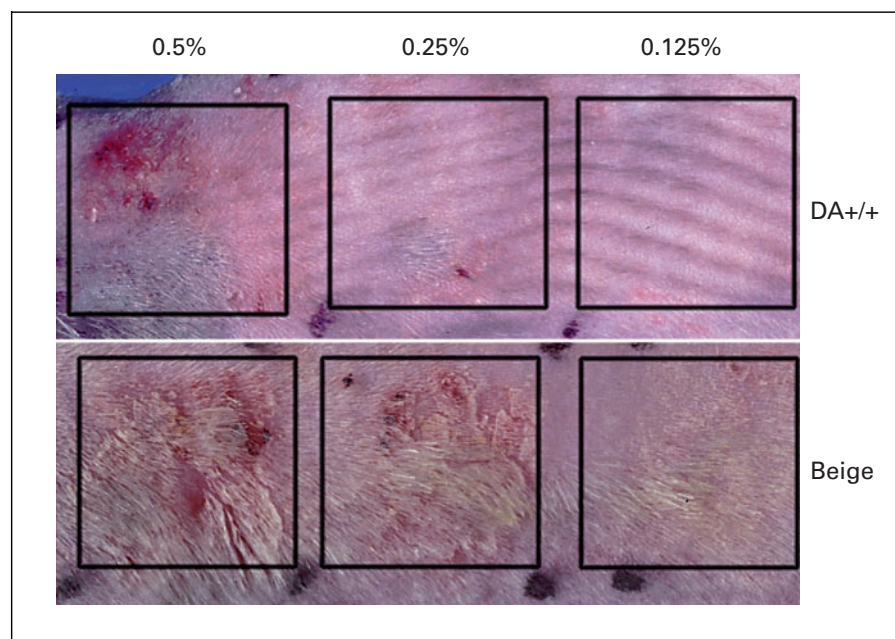
Fig. 5. Plasma level of total IgE in beige rats. Plasma levels of total IgE reach a low point at 4 and 8 weeks, increase slightly at 13 weeks and are markedly elevated from 16 weeks on, correlating with the severity of the skin lesions. That high level is maintained until 52 weeks. Each symbol represents the mean $\pm$ SD.

Discussion

Our results show that the skin lesions of beige rats are characterized by mixed responses to Th1 and Th2, and that type I allergy and high-contact sensitivity may initiate and accelerate them.

The characteristic histopathological changes in the acute lesions of dermatitis were epidermal hyperplasia and inflammatory cell infiltration of the dermis. Inflammatory cells included numerous CD3+, CD4+, ED1+, ED2+ and I-A+ mononuclear cells and eosinophils. In addition, scratching or self-trauma induced erosion and ulceration with crust formation. The skin lesions of humans with acute AD consisted of eczema with dermal inflammatory cell infiltration comprised of CD3+, CD4+, CD45RO+ and MHC class II+ mononuclear cells [24], confirming that acute lesions in the beige rat were similar to those of acute human AD. In chronic lesions of this rat, extensive dermal fibrosis was observed, but inflammatory cell infiltration was diminished. In the chronic lesions of human AD, the skin was lichenified with dendritic cells, macrophages and eosinophil infiltrations. T cells remained present, but in smaller numbers than those in acute lesions [5]. In addition, self-traumatic lesions also occurred, which consisted of focal epidermal necrosis, focal erosion above a sparse infiltration of inflammatory cells and bundles of degenerated collagen within a formidable crust [3]. Between beige rat and human AD, acanthosis and dermal fibrosis were similar, but infiltrated cell

Fig. 6. Maximization test of DA+/+ and beige rats 24 h after the challenge of DNCB. In beige rats, 0.5% DNCB shows a markedly positive response (score 3) while 0.25% DNCB shows a moderate response (score 2). However, in DA+/+ rats, only 0.5% DNCB produces a moderate response (score 2).



types were different. In particular, there was no increase in the numbers of dermal and epidermal antigen-presenting cells in chronic lesions of the beige rat. Thus, the histomorphological characteristics of spontaneous skin lesions in beige rats in some respects resembled those in the acute skin lesions of human AD, but differed from those in the chronic skin lesions of human AD.

Numerous mast cells were degranulated in lesions in the acute phase, but such cells decreased in number with progression of the inflammatory process, indicating that the degranulation of mast cells is related to the development and progression of acute dermal lesions. Mast cells of beige rats have been shown to release excessive amounts of histamine via the stimulation of IgE [25]. The elevation of serum IgE in human AD has been associated with the extent of the dermatitis [26]. A rather similar association has been observed in beige rats. Therefore, the exacerbation of dermal lesions was thought to have been initiated by the release of mediators after cross-linking of IgE on mast cells and to have progressed by pruritus that provoked scratching. The number of mast cells was unchanged, regardless of the stage of the lesions. In addition, some mast cells were degranulated even in chronic lesions, indicating that the degranulation of mast cells in chronic lesions repeatedly induced acute lesions.

The elevation of serum IgE was accompanied by the expression of IL-4 in the skin and lymph nodes of beige rats. As IL-4 induced IgE isotype switching and produc-

tion on B cells, thus leading to humoral immunity [2], the elevated serum IgE level of beige rats was thought to be induced by IL-4. IL-4 of the lymph nodes could be detected from the acute to the chronic stage, but that of the skin could only be detected at the acute stage. However, serum IgE maintained a high level from the acute to the chronic stage. Thus, IL-4 originated from lymph nodes rather than the skin, suggesting its influence on serum IgE levels.

PCA is a method for studying the mechanisms of immediate (type I) hypersensitivity [21, 27]. As beige rats showed high susceptibility to the PCA reaction, they had the capability of readily inducing IgE-mediated type I allergy. A maximization test is a method for detecting sensitization or hypersensitivity reactions, which include dermal cell-mediated type reactions [22]. Beige rats showed a high susceptibility to the maximization test, suggesting high skin contact sensitivity. As the high positivity of a patch test with house dust mites was found in human AD, contact hypersensitivity also played an important role in the progression of AD [14, 28]. Taken together, type I allergy and high contact sensitivity may be involved in the initiation and acceleration of dermatitis in beige rats.

Both Th2 cytokine IL-4 and Th1 cytokine IFN- γ expressions of acute lesions in beige rats were thought to constitute a mixed reaction of Th1 and Th2 [2, 6, 29]. In human acute AD, overexpression of the Th2 cytokines IL-

4 and IL-5 and the reduced production of Th1 cytokine IFN- γ were observed, indicating an augmentation of the Th2 response [4, 30–33]. However, IFN- γ mRNA was found to be highly expressed in chronic lesions of human AD [6, 7]. After successful therapy for human AD, the increased IFN- γ mRNA expression (but not the increased IL-4 mRNA expression) was significantly downregulated [7]. Unlike human AD patients, beige rats showed a high expression of IFN- γ in acute skin lesions and the absence of a shift from the Th2 to Th1 cytokine pattern during the chronification. Therefore, the expression pattern of cytokine in the beige rat was different from that in human AD.

RANTES expression on the skin of beige rats was detected in intact skin without eosinophil infiltration. RANTES is a chemoattractant for eosinophil and memory T cells recruited from the circulation to the inflammatory area [10, 34]. Thus, RANTES expression in the intact skin of beige rats may induce eosinophil recruitment prior to any pathological changes in the skin. RANTES in acute skin lesions was detected together with eosinophil and lymphocyte infiltration, and its expression was weakened with a decrease in the number of eosinophils and lymphocytes, indicating that the expression of RANTES was correlated with the levels of eosinophil and lymphocyte infiltration. RANTES mRNA and protein were detected and shown to markedly increase in severe human AD [11, 12]. Taken together, the expression patterns of RANTES in beige rats were also similar to those in human AD.

A spontaneous murine AD model, the NC/Nga mouse, has been reported. When NC/Nga mice were maintained

under conventional conditions, they showed clinical and histopathological changes, serum IgE levels and skin barrier changes similar to those in human AD [16, 18, 35]. One of the causes of dermatitis in NC/Nga mice was thought to be the parasitism of fur mites, since many mites were found in the areas with skin eruptions, but few in the areas without them. Moreover, no skin lesions occurred under specific pathogen-free conditions [36]. Beige rats also had fur mites on their skin. Clinical conditions, serum IgE and pathological lesions of beige rats were quite similar to those in NC/Nga mice. Fur mites were few and were not seen to directly induce distinctive inflammatory changes upon histopathological analysis [20]. In addition, they were not recognized as a significant pathogen in rats [37]. Therefore, an IgE-mediated allergic reaction to the mites might have contributed to the development of skin lesions, though the precise underlying mechanism was not clear. Thus, further investigations are needed to confirm the mechanism of dermatitis.

In conclusion, our results show that some of the skin lesions of beige rats are morphologically similar to AD, being characterized by the inflammatory cell composition in the acute phase, and increased IgE and RANTES levels. However, the inflammatory process and cytokine expression pattern are different from those in human AD.

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