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1. Organism

Heterogeneity of mast cells in mastocytosis and inhibitory effect of ketotifen and ranitidine on indolent systemic mastocytosis

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Background: Mastocytosis is a disorder of mast cell proliferation that occurs in both cutaneous and systemic forms. The most frequent site is the skin.

Objective: The mast cell subtype of two patients with mastocytosis was investigated.

Methods: Immunohistochemical studies were performed on the skin or gastric mucosa or both of the two patients. Blood and urine levels of various mediators were measured for one patient.

Results: Mast cells containing tryptase and chymase were the only type seen in the skin lesions of an 11-month-old boy with urticaria pigmentosa. Mast cells containing tryptase were predominant in lesions of the skin and gastric mucosa of a 41-year-old man with indolent systemic mastocytosis. However, mast cells containing tryptase and chymase were predominant in the nonlesional and the normal skin of this patient. Tryptase-positive cells were more numerous in lesional skin than nonlesional skin and normal skin. Elevated blood and urine levels of various mediators were decreased

by means of combination therapy with ketotifen and ranitidine.

Conclusion: In indolent systemic mastocytosis, mast cell dynamics involve only cells containing tryptase. Release of mediators from mast cells may be inhibited by means of combination therapy with histamine H1 and H2 receptor antagonists. (*J Allergy Clin Immunol* 1997;100:S25-32.)

Key words: Mast cell, tryptase, chymase, urticaria pigmentosa, systemic mastocytosis

Mastocytosis is a disorder of mast cell proliferation that occurs in both cutaneous and systemic forms. The most frequent site of involvement among patients with either form of mastocytosis is the skin, which can have a variety of lesions, including mastocytoma, urticaria pigmentosa, diffuse and erythrodermic mastocytosis, and telangiectasia macularis eruptiva perstans.¹

Mastocytomas usually appear within the first 3 months of life.² They are rare among adults because the lesions often resolve spontaneously. The most common cutaneous lesions among patients with mastocytosis are the red-brown macules, papules, and plaques of urticaria pigmentosa. When urticaria pigmentosa appears in infancy or childhood, more than half of patients experience spontaneous resolution by adolescence, and only lightly pigmented macules persist among the others.³

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Abbreviations used

EDTA:	Ethylenediamine tetraacetic acid
EIA:	Enzyme immunoassay
HPLC:	High-performance liquid chromatography
HPTLC:	High-performance thin-layer chromatography
HTMP:	4-Hydroxy-2,2,6,6-tetramethylpiperidine-1-oxyl
LT:	Leukotriene
MC _T cell:	Mast cell containing tryptase
MC _{TC} cell:	Mast cell containing tryptase and chymase
PG:	Prostaglandin
RIA:	Radioimmunoassay
TTBS buffer:	Buffer containing Tris and Tween-20

Mastocytosis is not a single well-defined disorder but is a group of diseases with heterogeneous manifestations and prognosis. The classification of mastocytosis originally proposed by Travis et al.⁴ and modified by Metcalfe⁵ is probably the most useful one available. Mastocytosis with a slower course is classified as indolent mastocytosis irrespective of systemic involvement.

Two subtypes of human mast cells are recognized on the basis of differences in the content of neutral proteases. Some mast cells contain tryptase but little or no chymase in their secretory granules (MC_T cells), and other mast cells contain substantial quantities of both tryptase and chymase (MC_{TC} cells).⁶ Immunohistochemical analysis of normal human tissues shows that MC_T cells are predominant in the gastrointestinal mucosa or alveolar walls, and MC_{TC} cells are predominant in the skin.⁷ The number of MC_T cells increases in inflammatory skin diseases.⁸⁻¹⁰ The results of studies by Algermissen et al.¹¹ suggest that disease-dependent changes in mast cell dynamics only involve MC_T cells among patients with cutaneous inflammation. We studied mast cell subtypes in two types of mastocytosis, urticaria pigmentosa and indolent systemic mastocytosis. We also studied the levels of various mediators for the patient with indolent systemic mastocytosis before and after he underwent combination therapy with histamine H1 and H2 receptor antagonists.

METHODS

Patients

The study involved two Japanese patients with mastocytosis who were treated at the department of dermatology at Gunma University Hospital. The first patient was an 11-month-old boy who was brought to us by his parents in June 1994 with red-brown papules on his torso. Darier's sign was present, and a diagnosis of urticaria pigmentosa was made. No specific treatment was needed, and the patient underwent follow-up examinations through May 1996. The second patient was a 41-year-old man who sought treatment in April 1996

with widespread brown-red maculae on the trunk and extremities and dyspepsia. The patient reported pruritus. Firm stroking of the skin revealed pronounced dermographism. The skin lesions had appeared in 1985 and had spread slowly over the years. Approximately 5 years later, the patient noticed episodes of flushing, sweating, irritability, and dyspepsia especially after exercise, hot baths, eating spicy foods, and drinking alcoholic beverages. These symptoms became more frequent over time, but no treatment had been attempted since 1985. After informed consent was obtained from the parents of the child and from the 41-year-old man, venous blood was collected from the man, and skin biopsy specimens were obtained from both patients. The study was approved by the institutional ethics committee of Gunma University School of Medicine.

Processing of biopsy tissues

In addition to biopsy specimens from the lesional skin, a biopsy specimen of nonlesional skin less than 3 cm away from the lesion was obtained from the 41-year-old man, and endoscopic biopsy specimens of his gastric lesions were obtained. As a control, biopsy specimens were obtained of normal-appearing skin that was surgically resected from corresponding sites on two patients without skin disease who served as controls. The tissues were immediately fixed at room temperature in Carnoy's fluid (60% ethanol, 30% chloroform, and 10% glacial acetic acid) for 24 hours and transferred to absolute ethanol. Tissues were embedded in paraffin, and sections 4 μ m thick were prepared. The sections were placed on glass slides coated with poly-L-lysine and air dried for at least 2 hours.

Immunohistochemistry

Monoclonal murine anti-tryptase and anti-chymase antibodies⁷ were provided by Prof. L. B. Schwartz, Virginia Commonwealth University, Richmond, Virginia. An immunohistochemical method was used for the detection of MC_T and MC_{TC} cells. In brief, sections were dewaxed in xylene and rehydrated with a graded alcohol series. Endogenous peroxidase was inhibited by means of incubation in methanol with 0.6% hydrogen peroxide for 30 minutes at room temperature. Serial sections were incubated in a humid chamber overnight at 4°C with a monoclonal murine anti-tryptase antibody (G3, 0.7 μ g/ml) diluted 1:400 in Tris buffer (50 mmol/L Tris HCl buffer at pH 7.6) containing 1% bovine serum albumin or with a monoclonal anti-chymase antibody (B7, 4 μ g/ml) diluted 1:200 in Tris buffer containing 1% bovine serum albumin. The sections were washed extensively in running buffer containing Tris and Tween-20 (TTBS) (50 mmol/L Tris HCl, 154 mmol/L NaCl, 0.05% Tween-20 at pH 7.4) at 4°C, followed by washing extensively in TTBS buffer for 15 minutes at room temperature (with three changes of buffer) and washing in distilled water for 5 minutes.

As described by Cordell et al.,¹² sections were incubated for 40 minutes in a humid chamber at room

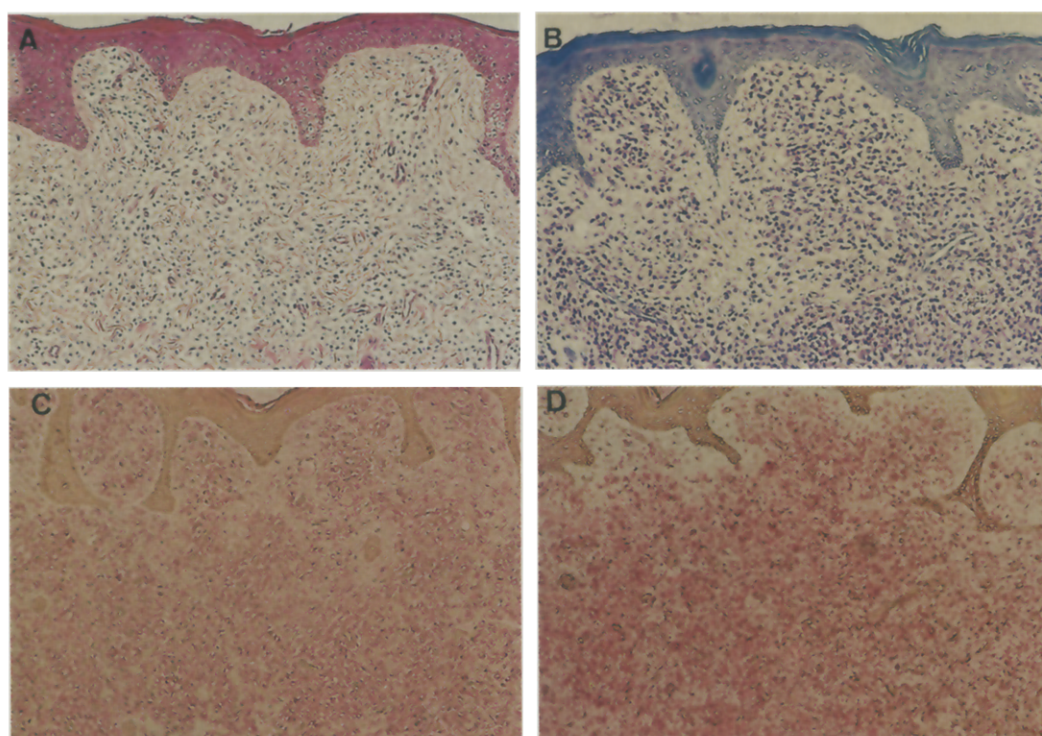


FIG. 1. Staining patterns of lesional skin ($\times 160$ magnification) from an 11-month-old boy with urticaria pigmentosa. **A**, Hematoxylin and eosin stain. **B**, Toluidine blue stain. **C**, Immunohistochemical stain for tryptase. **D**, Immunohistochemical stain for chymase.

temperature with a rabbit anti-mouse immunoglobulin antibody (Dako, Carpinteria, Calif.) diluted 1:100 with Tris buffer followed by washing in TTBS buffer for 15 minutes at room temperature (with three changes of buffer) and washing in distilled water for 5 minutes. Sections were incubated for 40 minutes in a humid chamber at room temperature with a monoclonal anti-alkaline phosphatase antibody conjugated to alkaline phosphatase (Dako) diluted 1:50 in Tris buffer. The sections were washed extensively in running TTBS buffer at 4° C then washed in TTBS buffer for 15 minutes at room temperature (with three changes of buffer) and washed in distilled water for 5 minutes. Color was developed for 10 minutes at room temperature with a fast red substrate that contained naphthol AS-MX phosphate (Dako) (100 mmol/L Tris HCl buffer at pH 8.2 containing naphthol phosphate, fast red, and levamisole). The sections were washed with distilled water, counterstained with Mayer's hematoxylin for 5 minutes, and washed in running water for 10 minutes and mounted in glycerol-gelatin (Sigma, St. Louis, Mo.). Photomicrographs were taken with a Nikon Microflex AFX-DX camera and an OPTIPHOT-2 microscope.

Mast cells were counted under a light microscope at $\times 160$ magnification by an investigator who was blinded to the clinical diagnosis of each patient. Mast cell concentrations were calculated according to the formula of Irani et al.,¹³ $\text{mast cells/mm}^3 = (\text{mast cells/mm}^2) \times [10^3/(9.5 + 4)]$, where 9.5 μm is the mean mast cell

TABLE I. Changes in blood and urine levels of mediators of a 41-year-old man with indolent systemic mastocytosis receiving combination therapy with H1 and H2 receptor antagonists

	Before therapy (April 1996)	After therapy (July 1996)	Normal range
Plasma histamine (ng/ml)	1.09	0.29	0.05–0.18
Urine methylhistamine (ng/mg Cr)	234.9	140.9	82.5–122.1
Serum tryptase (ng/ml)	5.7	<2.0	<2.0
Plasma PGE ₂ (pg/ml)	21.0	27.0	4.3–79.9
Plasma PGD ₂ (pg/ml)	19.5	75.4	25.0–43.0
Plasma 6ketoPGF _{1α} (pg/ml)	10.0	8.0	2.5–23.8
Plasma thromboxane B ₂ (pg/ml)	40.0	ND	2.1–61.0
Plasma LTB ₄ (pg/ml)	139.0	31.2	61.9–178.0
Plasma LTC ₄ (pg/ml)	80.5	39.4	<13.0
Urine LTE ₄ (pg/ml Cr)	541.5	332.6	<114.0

PG, Prostaglandin; LT, Leukotriene; Cr, Creatinine.

diameter¹⁴ and 4 μm is the section thickness. Mast cell concentrations in the dermis to a depth of 454 μm immediately below the superficial level were counted as described elsewhere.¹³ Mast cells in the periappendageal areas (sweat ducts and hair follicles) were not counted, because it has been reported that these cells normally

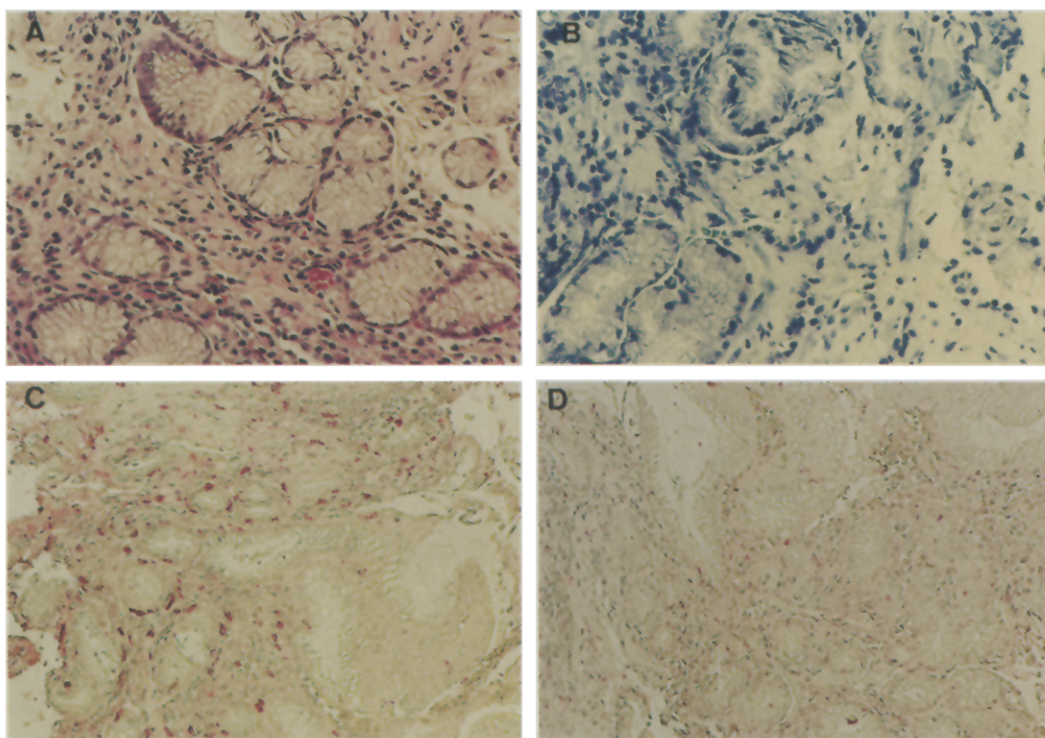


FIG. 2. Staining patterns of gastric mucosa ($\times 160$ magnification) from a 41-year-old man with indolent systemic mastocytosis. **A**, Hematoxylin and eosin stain. **B**, Toluidine blue stain. **C**, Immunohistochemical stain for tryptase. **D**, Immunohistochemical stain for chymase.

accumulate in the loose connective tissue around the skin appendages.¹¹ Tissue sections were stained by means of routine hematoxylin and eosin processing or with toluidine blue (pH 0.5) as described elsewhere.¹⁵

Assay of chemical mediators

Plasma histamine and urine *N*-methylhistamine levels were determined by means of radioimmunoassay (RIA) (Immunotech, Marseilles, France) as described by McBride et al.¹⁶ and by means of double antibody RIA (Pharmacia, Uppsala, Sweden) as described by Stephan et al.¹⁷ Serum tryptase level was determined with a tryptase RIA kit (Pharmacia) as described by Enander et al.¹⁸ Urinary creatinine level was determined with an L-type creatinine-F assay system (Wako Pure Chemical, Osaka, Japan). Urine levels of mediators were expressed per milligram of creatinine to correct for variations in urine output.

For measurement of leukotriene C_4 (LTC_4), 2 ml of whole blood treated with heparin was mixed with 8 ml of cold ethanol and centrifuged at 2000g for 15 minutes at 4° C, after which the supernatant was stored at -20° C until assay. Then 8.5 ml of sample was mixed with 8 ml of sodium phosphate buffer (pH 7.4) and 24 ml of dichloromethane. After centrifugation at 2000g for 10 minutes at 4° C, the upper water layer was loaded onto a Sep-Pak C18 cartridge (Waters, Milford, Mass.) conditioned with 10 ml of methanol and 10 ml of distilled water. After the cartridge was washed with 3 ml of 60%

methanol containing 0.1% acetic acid, the LTC_4 fraction was eluted with 4 ml of 80% methanol containing 0.1% acetic acid. The eluate was dried under nitrogen gas at 37° C and assayed with a LTC_4 [^3H] monoclonal antibody system (Amersham, Buckinghamshire, UK).

For measurement of LTE_4 , urine was mixed with buffer A (50 mmol/L sodium phosphate buffer at pH 7.4 containing 0.9% NaCl, 0.5 mmol/L ethylenediamine tetraacetic acid [EDTA] and 1 mol/L 4-hydroxy-2,2,6,6-tetramethylpiperidine-1-oxyl [HTMP]) at a 4:1 volume and stored at -20° C until assay. The sample was centrifuged at 2000g for 10 minutes at room temperature to remove particles. Then 10 ml of supernatant spiked with 3000 disintegrations per minute (dpm) of [^3H] LTE_4 as an internal standard for the assessment of recovery was loaded onto a Sep-Pak C18 cartridge (Waters) conditioned with 10 ml of methanol and 10 ml of distilled water. The cartridge was washed with 10 ml of distilled water and 3 ml of 40% methanol, and leukotrienes were eluted with 4 ml of 80% methanol. The eluate was evaporated to dryness under nitrogen gas at 37° C and reconstituted with 0.3 ml of high-performance liquid chromatography (HPLC) mobile phase (acetonitrile/methanol/0.05% acetic acid, pH 5.2, adjusted with ammonium hydroxide, 33:15:54, vol/vol/vol) before injection into a reversed-phase HPLC system. Separation was performed on a TSK-ODS-120T column (5 μ m, 4.6 mm $\times$ 250 mm; Tosoh, Tokyo, Japan) at a rate of 1 ml/min with isocratic elution. Fractions of 5 ml each at

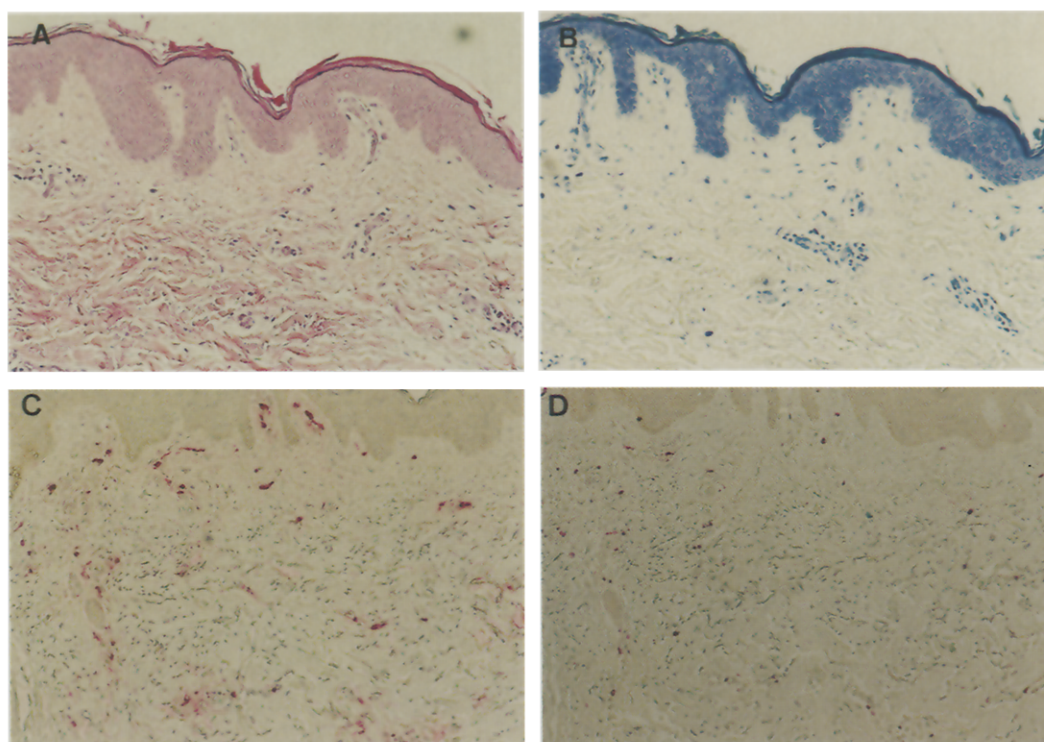


FIG. 3. Staining patterns of nonlesional skin ($\times 160$ magnification) from the patient with indolent systemic mastocytosis. **A**, Hematoxylin and eosin stain. **B**, Toluidine blue stain. **C**, Immunohistochemical stain for tryptase. **D**, Immunohistochemical stain for chymase.

a retention time were collected and evaporated to dryness under nitrogen gas at 37°C . The dried samples were reconstituted with 1 ml of assay buffer, and 0.3 ml of the resulting sample was used to detect ^3H radioactivity for estimation of the recovery. Assays were performed with an $\text{LTC}_4/\text{LTD}_4/\text{LTE}_4$ [^3H] assay system (Amersham).

For measurement of LTB_4 , 2 ml of whole blood treated with heparin was mixed with 8 ml of cold ethanol and centrifuged at 2000g for 15 minutes at 4°C after which supernatant was stored at -20°C until assay. Five milliliters of the supernatant spiked with 3000 dpm of [^3H] LTB_4 as an internal standard for the estimation of recovery was mixed with 15 ml of distilled water and loaded onto a Sep-Pak tC2 plus cartridge (Waters) conditioned with 4 ml of ethyl acetate, 4 ml of methanol, and 4 ml of distilled water. The cartridges were washed with 10 ml of distilled water, 4 ml of 10% ethanol, and 4 ml of hexane. Fractions containing LTB_4 were eluted with 3 ml of ethyl acetate, and the eluates were evaporated to dryness under nitrogen gas at 37°C . The dried samples were reconstituted with 0.5 ml of assay buffer, and 0.1 ml of each sample was used to determine ^3H radioactivity for the estimation of recovery. Assays were performed with an LTB_4 enzyme immunoassay (EIA) system (Amersham).

For measurement of prostaglandin D_2 (PGD_2), PGE_2 , 6-keto $\text{PGF}_{1\alpha}$, and thromboxane B_2 , 2 ml of whole blood was collected into a glass syringe containing EDTA and

indomethacin. After centrifugation at 2000g for 15 minutes at 4°C , the plasma was stored at -20°C until assay. For PGD_2 , 1 ml of sample was mixed with 2 ml of 10% acetic acid and applied to a C2 column (Amersham) conditioned with 10 ml of methanol and 10 ml of distilled water. The column was washed with 3 ml of distilled water, 3 ml of 10% ethanol, and 3 ml of hexane. PGD_2 fractions were eluted with 3 ml of methylformate, and the eluates were dried under nitrogen gas at 37°C . Assays were performed with a PGD_2 [^3H] assay system (Amersham).

For PGE_2 , 1 ml of sample was mixed with 3 ml of solvent A (0.1 N hydrochloric acid/ethyl acetate/2-propanol, 1:3:3, vol/vol/vol), 3 ml of distilled water, and 2 ml of ethyl acetate and centrifuged at 2000g for 10 minutes at 4°C . The upper phase was collected and dried under nitrogen gas at 37°C , dissolved in 2 ml of solvent D (ethyl acetate/benzene, 2:3, vol/vol), and applied to a silica gel column (Wako-gel Q-63, Wako Pure Chemical). The column was washed with 7 ml of solvent D, and the target fraction was eluted with 12 ml of solvent E (methanol/ethyl acetate/benzene, 0.1:2:3, vol/vol/vol). The eluate was dried under nitrogen gas at 37°C and assayed with a PGE_2 [^{125}I] assay system (Amersham).

For the assay of 6-keto $\text{PGF}_{1\alpha}$ and thromboxane B_2 , 1 ml of sample was mixed with 3 ml of solvent A, 3 ml of distilled water, and 2 ml of ethyl acetate and centrifuged at 2000g for 10 minutes at 4°C . The upper phase was collected and dried under nitrogen gas at 37°C , dis-

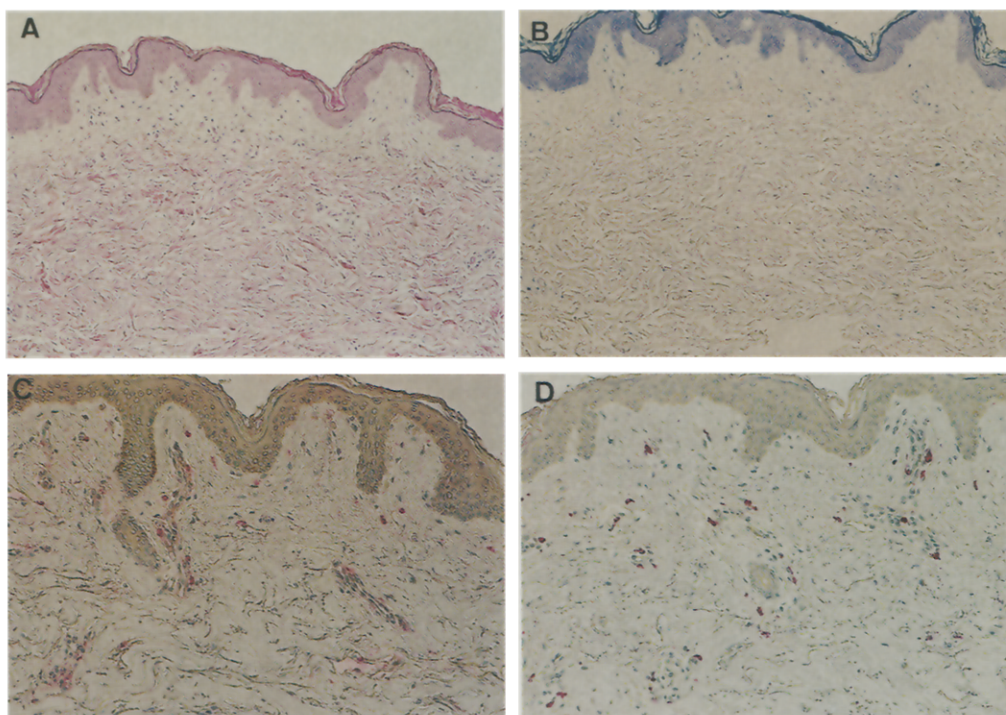


FIG. 4. Staining patterns of lesional skin ($\times 160$ magnification) from the patient with indolent systemic mastocytosis. **A**, Hematoxylin and eosin stain. **B**, Toluidine blue stain. **C**, Immunohistochemical stain for tryptase. **D**, Immunohistochemical stain for chymase.

solved in 2 ml of solvent B (ethyl acetate/benzene, 1:9, vol/vol), and applied to a silica gel column (Wako-gel Q-63). The column was washed with 4 ml of solvent B, and the target fraction was eluted with 4 ml of solvent C (methanol/ethyl acetate/benzene, 1:2:3, vol/vol/vol). The eluate was dried under nitrogen gas at 37°C , dissolved in 0.2 ml of ethanol, and applied to a high-performance thin-layer chromatography (HPTLC) glass plate coated with silica gel (HPTLC silica gel 60 plate, Merck, Rahway, NJ). The plate was developed twice with the mobile phase (ethyl acetate/isooctane/acetic acid/distilled water, 330:150:60:300, vol/vol/vol/vol). The zones corresponding to 6-ketoPGF $_{1\alpha}$ and thromboxane B $_2$ were localized by means of parallel development of 2 μg of standards for each mediator, which were identified with iodine vapor. The zones containing 6-keto prostaglandin F $_{1\alpha}$ and thromboxane B $_2$ were scraped off, mixed with 1 ml of 0.9% sodium chloride (pH 3.0) and 2 ml of ethyl acetate, and centrifuged at 2000g for 10 minutes at 4°C . The upper phase was collected in a siliconized glass tube, liquid-liquid extraction was performed twice, and the tubes were dried under nitrogen gas at 37°C . Assays for 6-keto prostaglandin F $_{1\alpha}$ and thromboxane B $_2$ were performed with a 6-ketoPGF $_{1\alpha}$ [^{125}I] assay system and a thromboxane B $_2$ [^{125}I] assay system (Amersham), respectively.

RESULTS

As shown in Fig. 1, MC $_{TC}$ cells were the predominant type of mast cell in the lesional skin from the 11-month-old boy with urticaria pigmentosa. There was no evi-

dence of malignant growth. The red-brown papules of this patient showed a tendency to resolve without treatment 2 years after diagnosis.

Table I shows the results of assays of various mediators in blood and urine specimens from the 41-year-old man. He had elevated levels of plasma histamine, serum tryptase, plasma LTC $_4$, urine methylhistamine, and urine LTE $_4$. At endoscopic examination, salmon pink patches were detected in the gastric antrum. Biopsy of these lesions showed an increase of metachromatic cells, MC $_T$ cells being the main type of mast cell in the gastric mucosa (Fig. 2). In contrast, MC $_{TC}$ cells were the main type of mast cell in nonlesional skin from this patient (Fig. 3), which was the same finding for healthy control skin from surgical specimens (data not shown). However, MC $_T$ cells were the main type of mast cell in the lesional skin of the adult patient (Fig. 4). The maximum number of mast cells in the dermis was found immediately below the dermoepidermal junction, the cells gradually falling to a minimum in the deeper layers of each specimen.

Mast cells were collected around the blood vessels and skin appendages (sweat ducts and hair follicles). The distribution of MC $_T$ cells and MC $_{TC}$ cells in the skin (excluding periappendageal areas) from the 41-year-old patient is shown in Table II. The average number of MC $_T$ cells in nonlesional skin was comparable with that in the corresponding control skin from surgical specimens. On the other hand, the number of MC $_T$ cells in the lesional skin of the patient was higher than in nonlesional skin or healthy skin. After a diagnosis of

indolent systemic mastocytosis was made, combination therapy with ketotifen (2 mg/day) and ranitidine (300 mg/day) was initiated. After 2 weeks of treatment, the gastrointestinal symptoms such as dyspepsia subsided, and the skin lesions showed some improvement. After 3 months of therapy, the symptoms were limited to the skin, and blood levels of mediators (plasma histamine, serum tryptase, and plasma LTC₄) and urine levels (methylhistamine and LTE₄) had decreased, even though the plasma PGD₂ level remained slightly high (Table I).

DISCUSSION

The most frequent site of involvement of mastocytosis is the skin. However, the type of mast cell present in the skin of patients with this disease is uncertain. Olafsson et al.¹⁹ reported an increase in the number of mast cells detected by means of metachromatic staining in skin fixed with Carnoy's fluid versus neutral-buffered formalin among patients with mastocytosis, suggesting the presence of mucosal mast cells in the skin. However, aldehyde blocking of dye binding is sometimes ambiguous as a method of differentiating MC_T cells from MC_{TC} cells. Ultrastructural studies with patients with urticaria pigmentosa and systemic mastocytosis^{20, 21} have shown mast cells with various stages of granule formation. It is difficult to differentiate the two types of mast cells in lesional skin. On the other hand, Irani et al.¹³ reported that only MC_{TC} cells exist in skin lesions, but the subjects were a combined group with systemic mastocytosis and urticaria pigmentosa. Because of the different clinical courses of systemic mastocytosis and urticaria pigmentosa, investigating the distribution of MC_T cells and MC_{TC} cells in separate groups would seem more reasonable.

In this study, we studied mast cell subtypes of a patient with urticaria pigmentosa and a patient with systemic mastocytosis. This study demonstrated that MC_{TC} cells accounted for mast cell proliferation in urticaria pigmentosa, whereas MC_T cells were found in the lesional skin of the patient with systemic mastocytosis. The presence of MC_T cells in actively inflamed skin lesions has been reported previously,⁸⁻¹¹ suggesting that only MC_T cells may be involved in cutaneous inflammation.

MC_T cells require helper T lymphocyte-derived factors for proliferation, maturation, or recruitment in gastrointestinal tissues, and a selective deficiency of MC_T cells is found in the gastrointestinal tissues of persons with congenital and acquired immunodeficiency diseases.²² However, a substantial number of T lymphocytes was not observed in the lesional skin of our patient with systemic mastocytosis. The exclusive presence of MC_T cells in his lesional skin suggests the possible involvement of local tissue factors in mast cell recruitment and differentiation.

This study showed that MC_{TC} cells were the only type found in normal skin, supporting the data reported by Irani et al.¹³ The number of mast cells in normal human dermis has been the subject of many investigations.

TABLE II. Average number of mast cells containing tryptase and mast cells containing tryptase and chymase in skin biopsy specimens from a patient with systemic mastocytosis and from healthy controls

Specimen	No. of mast cells per cubic millimeter	
	Containing tryptase	Containing tryptase and chymase
Healthy control skin	2919	3092
Nonlesional skin from the patient	2649	2621
Lesional skin from the patient	4907	1216

However, comparison of the results obtained from various studies is difficult because of variations in factors such as the depth of the skin examined, the thickness of the sections, the fixation technique, the staining technique, and the method of counting. In this study, we investigated the number of mast cells in the dermis using a modification of the mapping technique reported by Irani et al.¹³ They reported that mast cells were most densely distributed in the zone immediately below the dermoepidermal junction and that cell numbers decreased gradually in the deeper layers of the dermis. These findings are consistent with those of our study. In the lesional skin of the patient with systemic mastocytosis, an increase in the average number of mast cells was observed compared with nonlesional skin from the same patient and with healthy skin.

The primary mechanism accounting for the increase of MC_T cells in the skin lesions of our patient with systemic mastocytosis could be proliferation of mast cells or mast cell progenitors within the lesions, attraction of circulating or tissue-derived mast cell progenitors to these sites, or overproduction of mast cell progenitors by hematopoietic tissues. The presence multiple lesions on our patient with systemic mastocytosis without any evidence of malignant growth suggest that primary clonal expansion may not be the underlying defect. It is possible that an increased number of mast cell progenitors are attracted to or are encouraged to proliferate at discrete sites by tissue-dependent signals. The signals might be cytokines with mast cell chemotactic and growth activities, such as stem cell factor,²³ or produced by fibroblasts²⁴ or keratinocytes.²⁵ In urticaria pigmentosa, mast cells multiplying in the skin may lose their proliferative activity over time because of other tissue-dependent signals. A fibroblast-derived factor has been reported to be involved in the maintenance and maturation of rodent connective tissue mast cells (analogous to human MC_{TC} cells).²⁶ An analogous factor in persons, if expressed in an age-dependent manner, could be vital to the pathogenesis of urticaria pigmentosa and systemic mastocytosis.

Antihistamines are still the preferred therapy for

systemic mastocytosis. Treatment with histamine H1 receptor antagonists has been reported to be effective among some patients. The addition of histamine H2 receptor antagonists is logical considering the expression of both H1 and H2 receptors in the human body. Such therapy has been advocated for patients with gastrointestinal and cutaneous manifestations of systemic mastocytosis.²⁷ However, controlled studies have shown that the newer histamine receptor antagonists are not effective in mastocytosis.²⁸ The value of disodium cromoglycate has been disputed since a double-blind clinical trial failed to show any beneficial effect of this agent on systemic mastocytosis.²⁹ Our study showed that combination therapy with histamine H1 and H2 receptor antagonists was effective against cutaneous and gastrointestinal manifestations of indolent systemic mastocytosis. Because the elevated blood and urine levels of various mediators decreased after therapy, we suggest that the response was attributable not only to an effect on histamine receptors but also to interference with mast cell degranulation. Our patient had indolent systemic mastocytosis, which may be why combination therapy with histamine H1 and H2 receptor antagonists was effective.

One report³⁰ indicated that interferon alfa-2b may be very effective in aggressive mastocytosis, but Longley et al.³¹ found that two patients with systemic mastocytosis showed no response to treatment with interferon alfa-2b. At present, there is no specific cure for systemic mastocytosis, so treatment is symptomatic and must be tailored to the patient. However, early diagnosis of systemic mastocytosis may allow provision of reasonable therapy directed at prevention of mast cell degranulation or blockage of the effects of degranulation on target organs.

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