

Large maculopapular cutaneous lesions are associated with favorable outcome in childhood-onset mastocytosis

Tim Wiechers, MD,^{a,*} Anja Rabenhorst, PhD,^{a,*} Tina Schick, MD,^{a,b} Liane M. Preussner, MD,^a Anja Förster, PhD,^a Peter Valent, MD,^c Hans-Peter Horny, MD,^d Karl Sotlar, MD,^d and Karin Hartmann, MD^{a,e} *Cologne, Munich, and Luebeck, Germany, and Vienna, Austria*

Background: Mastocytosis, characterized by pathologic accumulation of mast cells, can manifest itself in adulthood or childhood. Pediatric patients usually have cutaneous mastocytosis (CM) with mast cell infiltrates limited to the skin and spontaneous improvement of skin lesions after several years. However, there are some patients with persistent disease resembling adulthood-onset mastocytosis.

Objective: The current classification of CM differentiates between 3 subforms. In clinical practice we noticed that different variants of these subforms might exist, particularly in patients with childhood-onset mastocytosis. Therefore, in the present study, we aimed to investigate whether specific cutaneous lesions in patients with childhood-onset mastocytosis are associated with other disease parameters.

Methods: We analyzed 144 patients with a disease onset of less than age 17 years using a systematic dermatologic approach.

Results: One hundred twenty-two patients presented with maculopapular cutaneous mastocytosis (MPCM), 12 patients presented with diffuse CM, and 10 patients presented with solitary mastocytoma of the skin. Patients with MPCM showed particularly heterogeneous cutaneous lesions and were therefore grouped into 3 variants presenting either with small lesions (MPCM-small, skin lesions <1 cm in diameter; n = 19), large lesions (MPCM-large, skin lesions ≥1 cm in diameter; n = 89), or atypical lesions (MPCM-other, n = 14). Patients with MPCM-large lesions, compared with those with MPCM-small lesions, were characterized by significantly lower tryptase levels, shorter disease duration, and earlier disease onset. In addition,

more patients with MPCM-large lesions exhibited spontaneous regression of cutaneous lesions.

Conclusion: Our data show that patients with MPCM-large lesions compared with those with MPCM-small lesions have a more favorable disease course and suggest exploring the size of cutaneous lesions as a prognostic parameter in childhood-onset MPCM. (*J Allergy Clin Immunol* 2015;■■■:■■■-■■■.)

Key words: Childhood-onset mastocytosis, cutaneous mastocytosis, KIT mutation, mast cell, mastocytosis, pediatric mastocytosis, prognosis, skin, tryptase, urticaria pigmentosa

Mastocytosis is characterized by pathologic accumulation of mast cells (MCs) in tissues.^{1,2} The organ most frequently affected is the skin, followed by bone marrow and the gastrointestinal tract. The current World Health Organization classification includes cutaneous mastocytosis (CM), which is defined by MC infiltrates confined to the skin, and various categories of systemic mastocytosis (SM), which are defined by accumulation of MCs in at least 1 extracutaneous organ, often combined with skin involvement.²⁻⁶ Because of the increased release of MC mediators, patients with mastocytosis can experience pruritus, flushing, headache, abdominal cramping, diarrhea, and anaphylaxis.

Onset of mastocytosis can occur in adulthood or childhood.⁷⁻¹⁴ Recent studies revealed several differences between adulthood- and childhood-onset mastocytosis.¹⁰ For example, adult patients usually carry the activating mutation *KIT* D816V located in exon 17 of the *KIT* gene, whereas children express diverse *KIT* mutations affecting either exon 8, 9, 10, 11, or 17.¹⁵⁻²² Most adults exhibit SM, whereas most pediatric patients have CM. Anaphylaxis is a frequent symptom in adults but only rarely concerns children.²³ Moreover, adult patients usually experience a chronic course, and children often show a transient course with spontaneous regression after several years.²⁴⁻³⁰

Patients with MC infiltrates in the skin exhibit heterogeneous cutaneous lesions.³¹⁻³⁷ The current classification of CM differentiates between 3 subforms, namely maculopapular cutaneous mastocytosis (MPCM)/urticaria pigmentosa, diffuse cutaneous mastocytosis (DCM), and solitary mastocytoma of the skin (mastocytoma).^{2,4,5} However, in clinical practice we noticed that different variants of these subforms might exist, particularly in childhood-onset mastocytosis. Therefore in the present study we sought to investigate whether specific cutaneous lesions in patients with childhood-onset mastocytosis are associated with other disease parameters. Using a systematic dermatologic approach, we found that patients with large MPCM lesions compared with those with small lesions are characterized by lower tryptase levels, shorter disease duration, and more frequent spontaneous improvement.

From ^athe Department of Dermatology, University of Cologne; ^bthe Department of Ophthalmology, University Hospital of Cologne; ^cthe Department of Internal Medicine I, Division of Hematology & Hemostaseology, Medical University of Vienna; ^dthe Institute of Pathology, Ludwig-Maximilians-Universität, Munich; and ^ethe Department of Dermatology, University of Luebeck.

*These authors contributed equally to this work.

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Corresponding author: Karin Hartmann, MD, Department of Dermatology, University of Luebeck, Ratzeburger Allee 160, 23538 Luebeck, Germany. E-mail: karin.hartmann@uksh.de.

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Abbreviations used

CM: Cutaneous mastocytosis
 DCM: Diffuse cutaneous mastocytosis
 MC: Mast cell
 MPCM: Maculopapular cutaneous mastocytosis
 SM: Systemic mastocytosis

METHODS**Patients**

Our study includes 144 patients with childhood-onset mastocytosis who regularly attended the Mastocytosis Clinic of the Department of Dermatology, University of Cologne, Cologne, Germany, between 1999 and 2012 (Table I). Inclusion criteria were the presence of CM lesions and a disease onset of less than age 17 years (mean $\pm$ SD, 18.8 ± 42.1 months; range, 0-192 months). At initial presentation, age ranged from 0 to 61 years (mean $\pm$ SD, 10.6 ± 14.2 years). Patients were given diagnoses and classified into different disease categories according to established criteria.^{2,4,5} Measurement of serum tryptase levels was performed at the patient's first visit by using a fluoroenzyme assay (ImmunoCAP Tryptase; Thermo Fisher Scientific, Uppsala, Sweden; mean $\pm$ SD, 23.6 ± 46.6 μ g/L; range, 1.4-293.0 μ g/L). In agreement with current recommendations, a bone marrow biopsy was only performed in adult patients.⁴

Characterization of CM lesions

CM lesions of all patients were assessed by an experienced dermatologist (K.H.) and a resident (T.W.) using photographic images that were routinely taken by photographers of the Photo Department, University Hospital Cologne, Cologne, Germany, at initial presentation and at follow-up visits after achieving written informed consent. Additionally, patients were asked to provide previous pictures of themselves (eg, from their beach holidays during childhood).

By defining the morphology and distribution of skin lesions, patients were first classified into 3 groups presenting with either MPCM, DCM, or mastocytoma (Fig 1). Cutaneous lesions of all patients with MPCM were then further defined as being small (skin lesions <1 cm in diameter), large (skin lesions $\geq$ 1 cm in diameter), or other (atypical lesions that could not be clearly described as being small or large) by defining diameter as the largest possible distance between 2 points along the border of the skin lesion. Moreover, MPCM lesions were categorized as being flat or elevated, showing a sharp or indistinct margination, round or polycyclic shape, and confluent or not confluent arrangement (see Fig E1 in this article's Online Repository at www.jacionline.org).

Mutational analysis

The presence of *KIT* codon 816 mutations was investigated in formalin-fixed and paraffin-embedded skin biopsy specimens, as previously described.³¹ Briefly, total genomic DNA was extracted from paraffin-embedded sections by using the QIAamp DNA Micro kit (Qiagen, Hilden, Germany). Total DNA was used for *KIT* mutation analyses performed by using melting point analysis of amplification products with the LightCycler System (Roche Molecular Systems, Mannheim, Germany). Amplicons to be genotyped were generated by means of nested PCR and locked nucleic acid-mediated PCR clamping to increase diagnostic sensitivity.

Histologic analysis

Skin biopsy specimens were fixed in 4% neutral-buffered formalin and embedded in paraffin. Immunohistochemical investigations were performed with antibodies against MC tryptase (1:1200; clone AA-1; Diagnostic BioSystems, Pleasanton, Calif) and CD25 (1:50; clone 4C9; Novocastra, Newcastle, United Kingdom) by using the avidin-biotin immunoperoxidase staining technique. In tryptase-stained sections MC density, MC localization

TABLE I. Clinical characteristics of patients with childhood-onset mastocytosis and skin lesions participating in the study

Parameters	All	MPCM	DCM	Mastocytoma
All, no. (%)	144 (100)	122 (84.7)	12 (8.3)	10 (6.9)
Sex				
No.	144	122	12	10
Male, no. (%)	71 (49.3)	59 (48.4)	6 (50.0)	6 (60.0)
Female, no. (%)	73 (50.7)	63 (51.6)	6 (50.0)	4 (40.0)
Disease onset (mo)				
No.	142	120	12	10
Mean $\pm$ SD	18.8 ± 42.1	21.6 ± 45.2	2.8 ± 2.3	5.2 ± 8.2
Range	0-192	0-192	0-6	0-25
Disease duration (y)				
No.	144	122	12	10
Mean $\pm$ SD	10.0 ± 12.5	9.5 ± 11.1	19.2 ± 23.4	5.1 ± 4.6
Range	0-71	0-70	1-71	0-13
Tryptase (μ g/L)*				
No.	109	93	11	5
Mean $\pm$ SD	23.6 ± 46.6	16.5 ± 35.8	90.2 ± 78.5	9.3 ± 4.7
Range	1.4-293.0	1.4-265.0	35.4-293.0	4.9-16.0

Statistically significant differences are shown in boldface.

*Tryptase: MPCM and DCM, $P < .0001$; DCM and mastocytoma, $P = .0022$.

within the dermis, MC infiltration pattern, MC morphology, and percentage of spindle-shaped MCs were determined.

Statistical analysis

Statistical analysis was performed by using GraphPad Prism software (version 5.01; GraphPad Software, La Jolla, Calif) with the nonparametric Mann-Whitney U test. Correlation analysis was performed with Spearman rank order correlation. A P value of less than .05 was considered statistically significant.

RESULTS**Study population**

To investigate whether specific CM lesions are associated with other disease parameters in patients with childhood-onset mastocytosis, we retrospectively analyzed 144 patients who presented with skin lesions and had mastocytosis onset at less than 17 years of age (Table I). Of these 144 patients, 122 (84.7%) were given a diagnosis of MPCM, 12 (8.3%) were given a diagnosis of DCM, and 10 (6.9%) were given a diagnosis of solitary mastocytoma (Fig 1). Serum tryptase levels at initial presentation were significantly increased in patients with DCM compared with those in patients with MPCM or mastocytoma (Table I; mean $\pm$ SD: MPCM, 16.5 ± 35.8 μ g/L; DCM, 90.2 ± 78.5 μ g/L; mastocytoma, 9.3 ± 4.7 μ g/L). There was no significant difference in sex, disease onset, and disease duration between those with MPCM, DCM, or mastocytoma.

Characteristics of cutaneous lesions in childhood-onset mastocytosis

Cutaneous lesions were next defined in more detail by using dermatologic terms of morphology and distribution (Fig 1 and Table II). Patients with DCM and mastocytoma showed rather homogeneous skin lesions typical for DCM and mastocytoma, respectively, whereas patients with MPCM presented with heterogeneous lesions, which differed from patient to patient in, for example, size, margination, and distribution (Fig 1).

Serum trypsin levels were compared between different subforms and variants of CM (Fig 2, A). Patients with MPCM-small lesions and DCM showed particularly high trypsin levels. Whereas trypsin levels in patients with MPCM-small lesions varied between normal and highly increased values (mean $\pm$ SD, 44.0 $\pm$ 70.0 μ g/L; range, 3.4-265.0 μ g/L), all

TABLE II. Morphologic characteristics and arrangement of skin lesions in patients with MPCM

Parameters	MPCM	Morphology of MPCM lesions									Arrangement of MPCM lesions	
		Size			Elevation		Margination		Shape		Confluence	
		Small, <1 cm in diameter	Large, ≥1 cm in diameter	Other, NA, atypical	Flat	Elevated	Sharp	Indistinct	Round	Polycyclic	Confluent	Not confluent
All, no. (%)	122 (100)	19 (15.6)	89 (72.9)	14 (11.5)	70 (57.4)	26 (21.3)	54 (44.3)	38 (31.1)	71 (58.2)	25 (20.5)	23 (18.9)	74 (60.7)
Sex												
No.	122	19	89	14	70	26	54	38	71	25	23	74
Male, no. (%)	59 (48.4)	5 (26.3)	51 (57.3)	3 (21.4)	35 (50.0)	14 (53.8)	28 (51.9)	21 (55.3)	40 (56.3)	9 (36.0)	11 (47.8)	38 (51.4)
Female, no. (%)	63 (51.6)	14 (73.7)	38 (42.7)	11 (78.6)	35 (50.0)	12 (46.2)	26 (48.1)	17 (44.7)	31 (43.7)	16 (64.0)	12 (52.2)	36 (48.6)
Disease onset (mo)*												
No.	120	19	87	14	70	24	52	38	71	23	23	72
Mean ± SD	21.6 ± 45.2	87.8 ± 76.4	4.7 ± 5.3	36.9 ± 43.3	21.9 ± 44.5	3.6 ± 5.2	14.0 ± 37.5	18.9 ± 40.4	19.5 ± 42.9	10.3 ± 24.7	17.7 ± 36.7	18.0 ± 40.8
Range	0-192	0-192	0-30	0-120	0-174	0-24	0-174	0-168	0-174	0-120	0-120	0-174
Disease duration (y)†												
No.	122	19	89	14	70	26	54	38	71	25	23	74
Mean ± SD	9.5 ± 11.1	24.7 ± 17.9	5.9 ± 4.8	11.4 ± 10.6	9.9 ± 10.8	5.7 ± 4.4	7.7 ± 8.6	9.8 ± 10.8	8.3 ± 8.6	10.2 ± 12.3	11.3 ± 11.2	8.2 ± 9.1
Range	0-70	2-70	0-22	0-22	0-58	0-15	0-58	1-37	0-37	1-58	1-37	0-58
Tryptase (μg/L)‡												
No.	93	19	62	12	56	17	40	29	54	19	21	53
Mean ± SD	16.5 ± 35.8	44.0 ± 70.0	7.1 ± 4.8	21.4 ± 27.3	15.6 ± 36.9	13.9 ± 15.7	9.6 ± 11.0	23.8 ± 50.2	16.7 ± 37.4	10.7 ± 15.1	34.2 ± 57.6	7.8 ± 6.4
Range	1.4-265.0	3.4-265.0	1.4-23.7	4.1-86.7	1.4-265.0	1.4-66.5	1.4-66.5	1.9-265.0	1.4-265.0	2.8-67.0	3.9-265.0	1.4-30.2

Statistically significant differences are shown in boldface.

NA, Not applicable.

*Disease onset: MPCM-small and MPCM-large, $P = .0003$; MPCM-large and MPCM-other, $P = .0028$; flat and elevated, $P = .0022$.

†Disease duration: MPCM-small and MPCM-large, $P < .0001$.

‡Tryptase: MPCM-small and MPCM-large, $P < .0001$; confluent and not confluent, $P < .0001$.

patients with DCM exhibited clearly increased levels ($>35 \mu\text{g/L}$) at initial presentation (mean ± SD, $90.2 \pm 78.5 \mu\text{g/L}$; range, 35.4-293.0 $\mu\text{g/L}$). On the other hand, all patients with MPCM-large lesions and mastocytoma had normal or only slightly increased tryptase levels (MPCM-large: mean ± SD, $7.1 \pm 4.8 \mu\text{g/L}$; range, 1.4-23.7 $\mu\text{g/L}$; mastocytoma: mean ± SD, $9.3 \pm 4.7 \mu\text{g/L}$; range, 4.9-16.0 $\mu\text{g/L}$).

Previous studies have demonstrated correlations between tryptase levels and MC numbers³⁸ and tryptase levels and the *KIT* D816V mutant allele burden in peripheral blood³⁹⁻⁴² and in adult patients between tryptase levels and the extent and density of skin lesions.³³ To examine whether tryptase levels also correlate with numbers of MPCM-large lesions in patients with childhood-onset mastocytosis, patients with MPCM-large lesions were grouped according to the number of cutaneous lesions (Fig 2, B). We observed a minor increase in tryptase levels in patients with more than 100 lesions (mean ± SD, $9.1 \pm 5.8 \mu\text{g/L}$) compared with levels in patients with only 2 to 50 cutaneous lesions (mean ± SD, $6.1 \pm 4.2 \mu\text{g/L}$).

Analyzing the 12 patients with DCM, of whom 11 had increased tryptase levels at initial presentation, in more detail (Fig 2, C, and Table III), we noticed that 6 patients had the rare subform of familial mastocytosis compared with 5 patients with sporadic disease and 1 patient with an unknown family history. In 4 patients with DCM, we were able to record the course of tryptase levels over 7 to 11 years (Fig 2, C). Of note, patients with sporadic DCM showed a gradual decrease in tryptase levels, whereas patients with familial DCM exhibited rather stable tryptase levels. At initial presentation, tryptase levels were significantly different between patients with sporadic (mean ± SD, $93.2 \pm 50.5 \mu\text{g/L}$) and familial (mean ± SD, $46.7 \pm 14.1 \mu\text{g/L}$) forms of the disease

(Table III). Moreover, all patients with sporadic DCM displayed a decrease in skin lesions after several years, whereas all patients with familial DCM had no decrease in cutaneous lesions. There was no difference in sex, disease onset, and duration between patients with sporadic and those with familial DCM.

Disease duration in different subforms of CM

We next investigated the duration of mastocytosis in the different subforms and variants of CM, defining disease duration as the time period from diagnosis to last contact with our mastocytosis clinic (Fig 3 and Table II). Disease duration was increased in patients with MPCM-small lesions and DCM compared with duration in those with the other subforms (Fig 3, A). In particular, patients with MPCM-small lesions had a significantly increased disease duration compared with those with MPCM-large lesions (MPCM-small lesions: mean ± SD, 24.7 ± 17.9 years; MPCM-large lesions: mean ± SD, 5.9 ± 4.8 years). Subdividing patients into groups with different disease durations (Fig 3, B), we also found that most patients with MPCM-small lesions had a long disease duration (<8 years: 1/19 patients, 5.3%; 8-14 years: 7/19 patients, 36.8%; and >14 years: 11/19 patients, 57.9%), whereas most patients with MPCM-large lesions had a short disease duration (<8 years: 61/89 patients, 68.5%; 8-14 years: 23/89 patients, 25.8%; and >14 years: 5/89 patients, 5.6%). In addition, disease duration was found to positively correlate with tryptase levels in all study patients (Fig 3, C) as well as in patients with MPCM (Fig 3, D) and patients with MPCM-small lesions (Fig 3, E).

Disease onset in different subforms of CM

Analyzing disease onset in patients with the different subforms of CM, we again observed a clear difference between

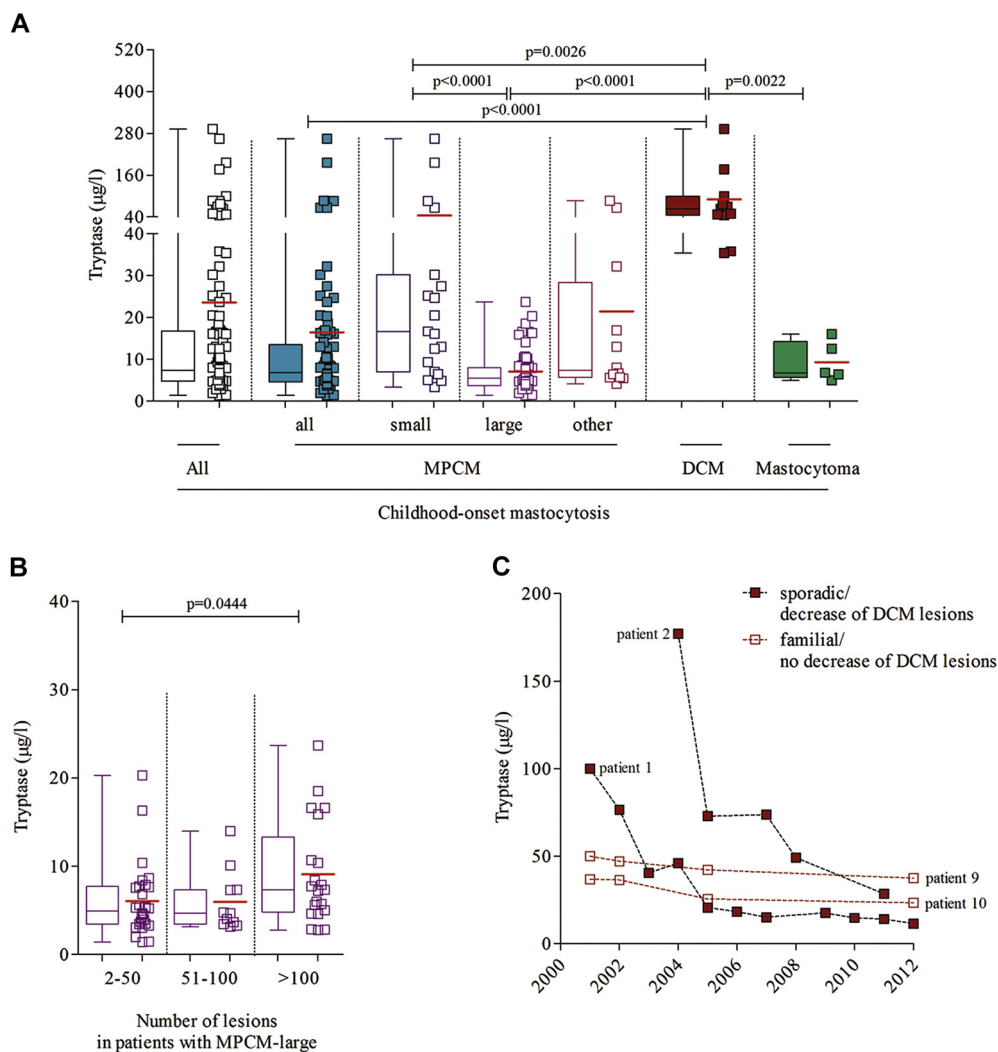


FIG 2. Serum tryptase levels differ between cutaneous subforms and variants. **A** and **B**, Tryptase levels were measured in patients with different cutaneous subforms (Fig 2, **A**) and in patients with MPCM-large lesions subdivided according to the number of skin lesions (Fig 2, **B**). Data are presented as box plots (minimum to maximum) with medians (left) and scatter dot plots with means (right). **C**, Representative course of tryptase levels over 7 to 11 years in 2 patients with sporadic DCM and decrease in cutaneous involvement (patient 1 and 2 of Table III; solid squares) and in 2 patients with familial DCM and no decrease in cutaneous involvement (patients 9 and 10 of Table III, open squares).

MPCM-small and MPCM-large lesions (Table II and see Fig E2 in this article's Online Repository at www.jacionline.org). Although most patients with MPCM-small lesions had disease onset after the age of 24 months (>24 months: 11/19 patients, 57.9%), most patients with MPCM-large lesions had onset at less than age 7 months (0 months [at birth]: 13/89 patients, 14.6%; 1-3 months: 33/89 patients, 37.1%; and 4-6 months: 29/89 patients, 32.6%). Also, all patients with DCM had disease onset at less than 7 months of age (Table III and Fig E2; 0 months [at birth]: 4/12 patients, 33.3%; 1-3 months: 4/12 patients, 33.3%; and 4-6 months: 4/12 patients, 33.3%).

Disease course in different subforms of CM

Cutaneous lesions are known to often resolve spontaneously in patients with childhood-onset mastocytosis. To investigate the course of skin lesions in the different subforms of CM,

we next recorded the number of patients with a decrease in cutaneous lesions over time (Fig 4), defining the decrease as either shrinking or fading of the lesions. Of 125 patients, 52 (41.6%) had decreasing skin lesions (Fig 4, **A**). In particular, among the patients with MPCM-large lesions and mastocytoma, more than half showed a decrease in cutaneous lesions (MPCM-large lesions, decrease: 36/70 patients, 51.4%; mastocytoma: 6/10 patients, 60.0%). In contrast, only a few patients with MPCM-small lesions had a decrease in lesions (MPCM-small, decrease: 3/19 patients, 15.8%). Representative examples of a patient with MPCM-large lesions who had prominent elevated lesions at the beginning of the disease that almost fully resolved over a period of 5 years (Fig 4, **B**, upper panel) and a patient with MPCM-small lesions who, on the other hand, had stable lesions over 9 years (Fig 4, **B**, lower panel) are depicted in Fig 4, **B**. These data suggest that spontaneous improvement preferentially occurs

TABLE III. Clinical characteristics of patients with DCM

No.	Occurrence	Family no.	Sex	Disease onset (mo)	Disease duration (y)	Tryptase ($\mu\text{g/L}$)*	Decrease/no decrease in DCM lesions
1	Sporadic	NA	Male	4	7	100.0	Decrease
2	Sporadic	NA	Male	6	5	177.0	Decrease
3	Sporadic	NA	Male	0	6	77.0	Decrease
4	Sporadic	NA	Male	0	8	49.1	Decrease
5	Sporadic	NA	Female	5	4	63.0	Decrease
Mean				3.0 $\pm$ 2.8	6.0 $\pm$ 1.6	93.2 $\pm$ 50.5	
$\pm$ SD							
Range				0-6	4-8	49.1-177.0	
6	Familial	Family 1	Male	6	1	ND	No decrease
7	Familial		Female	3	4	70.0	No decrease
8	Familial	Family 2	Female	3	71	45.0	No decrease
9	Familial		Female	3	48	47.1	No decrease
10	Familial		Female	0	18	35.8	No decrease
11	Familial		Female	3	8	35.4	No decrease
Mean				3.0 $\pm$ 1.9	25.0 $\pm$ 28.3	46.7 $\pm$ 14.1	
$\pm$ SD							
Range				0-6	1-71	35.4-70.0	
12	Unknown	NA	Male	0	50	293.0	No decrease

Statistically significant differences are shown in boldface.

NA, Not applicable; ND, not detected.

*Tryptase: sporadic and familial, $P = .0317$.

in patients with MPCM-large lesions but not in patients with MPCM-small lesions.

Disease duration until the start of decrease was recorded in 32 patients with MPCM-large lesions to examine at which age the decrease in cutaneous lesions develops (Fig 4, C). In most patients we observed decreasing lesions starting after a disease duration of 5 to 6 years (5-6 years: 13/32 patients, 40.6%).

Mutational and histologic characteristics in different subforms of CM

Next, skin sections of 30 patients were analyzed for the presence of *KIT* mutations in codon 816 and histologic characteristics (see Fig E3 in this article's Online Repository at www.jacionline.org). Of 28 patients in whom mutational analysis could be performed successfully, 9 (32.1%) expressed the mutation *KIT* D816V, 1 (3.6%) expressed the mutation *KIT* D816Y, and 18 (64.3%) had the wild-type codon 816. In this limited number of patients, there was no clear association between these molecular findings and histologic characteristics, such as MC density, MC localization within the dermis, the presence of MC aggregates, MC infiltration pattern, MC morphology, and the presence of spindle-shaped MCs (see Fig E3). Of note, most patients with the mutation *KIT* D816V did not express CD25 in the skin. Furthermore, the 3 patients with DCM, who all exhibited wild-type *KIT* in codon 816, showed a similar histologic pattern with the presence of MC aggregates, high MC density, round MCs (no spindle-shaped MCs), and high expression of CD25.

DISCUSSION

In the present study we used a systematic dermatologic approach to explore whether specific cutaneous lesions in patients with childhood-onset mastocytosis are associated with other disease parameters. In the group of patients with MPCM, we found that those with large lesions compared with those with small lesions are characterized by lower tryptase levels, shorter disease duration, and earlier disease onset. Furthermore, more

patients with MPCM-large lesions exhibited a spontaneous decrease in their skin lesions. These data suggest that patients with MPCM-large lesions have a more favorable outcome of the disease.

There are only few studies addressing cutaneous lesions in childhood-onset mastocytosis.^{25-30,36,43,44} Several of these previous studies also used different classifications and can therefore only partially be compared with our study.^{28,44} In accordance with the results of 3 studies,^{25,26,43} most patients of our study population presented with MPCM (84.7%), followed by DCM (8.3%) and solitary mastocytoma (6.9%, Table I). In contrast, Hannaford and Rogers²⁹ reported on a larger percentage of patients with mastocytoma. This difference might be due to the fact that our mastocytosis clinic is a specialized referral center.

Our study population had a male/female ratio of 1:1 (Table I), which is in line with previous investigations reporting on an equal sex distribution,^{11,13} although also male^{26,27} and female¹⁸ predominance have been described.

To our knowledge, this is one of the first studies that systematically investigates macroscopic features of MPCM lesions in patients with childhood-onset mastocytosis (Table II).⁴⁵ Analyzing the size, elevation, margination, shape, and confluence, we noticed that in particular the size of MPCM lesions affected several disease parameters. Similarly, other studies have also reported on larger skin lesions in pediatric compared with adult patients.^{33,36} For example, Brockow et al³³ observed that MPCM lesions in children had greater mean and maximum diameters compared with those in adults.

Patients with MPCM-large lesions had significantly lower serum tryptase levels compared with those with MPCM-small lesions (Fig 2, A, and Table II). Because tryptase levels of less than 20 $\mu\text{g/L}$ are usually found in patients with CM and levels of greater than 20 $\mu\text{g/L}$ are usually found in patients with SM,^{32,46} this finding suggests that patients with MPCM-large lesions mostly reflects CM, whereas patients with MPCM-small lesions might often be associated with SM.³¹

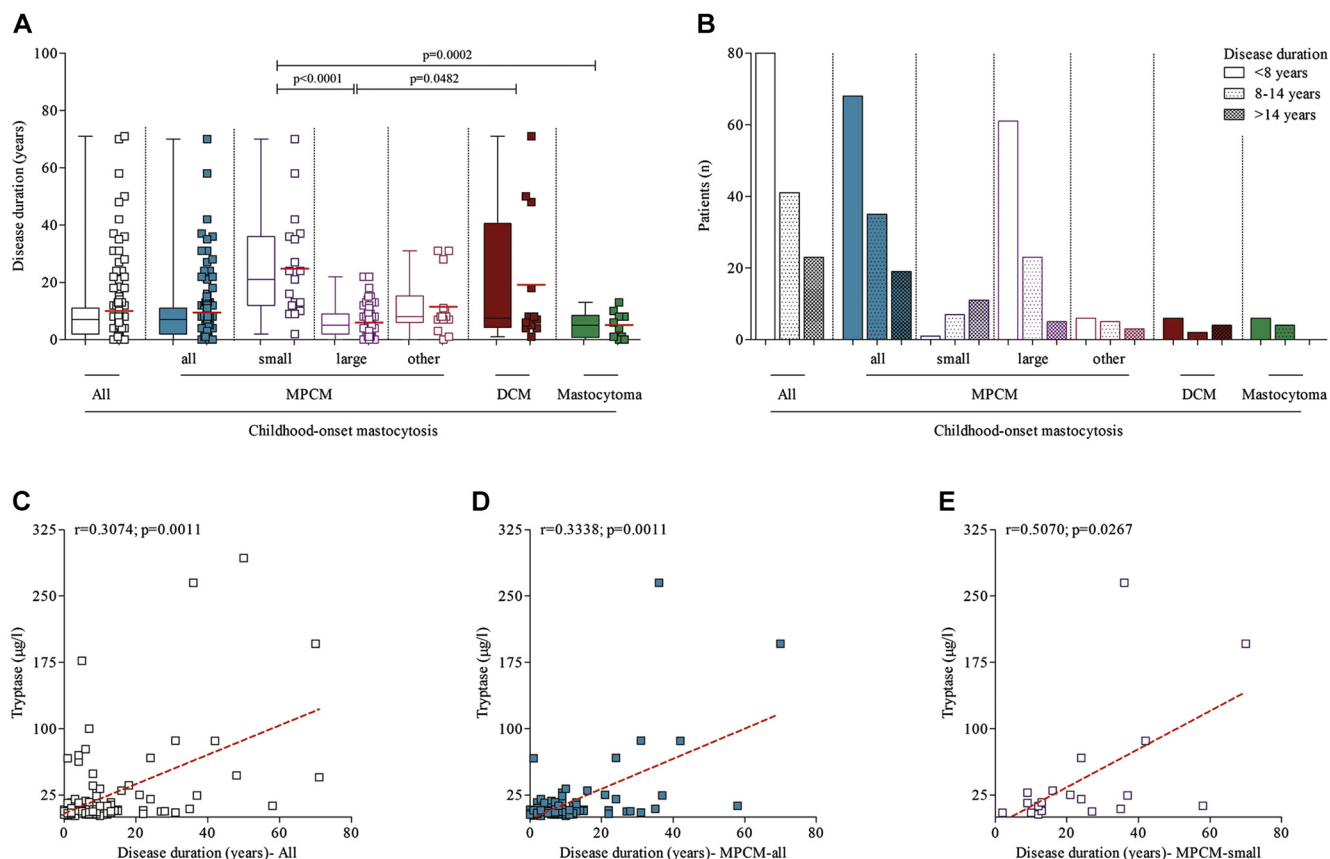


FIG 3. Disease duration differs between cutaneous subforms and correlates with tryptase levels. **A**, Disease duration was recorded in patients with different cutaneous subforms. Data are presented as *box plots* (minimum to maximum) with medians (*left*) and *scatter dot plots* with means (*right*). **B**, Number of patients grouped according to disease duration. **C-E**, Tryptase levels (*y-axis*) positively correlate with disease duration (*x-axis*) in all patients with childhood-onset mastocytosis (Fig 3, *C*), all patients with MPCM (Fig 3, *D*), and patients with MPCM-small lesions (Fig 2, *E*).

Brockow et al³³ have demonstrated that the extent and density of skin lesions correlate with tryptase levels in adults; however, they did not find this correlation in children. In contrast, we observed slightly increased tryptase levels in children with MPCM-large lesions with more than 100 skin lesions compared with children with only 2 to 50 lesions (Fig 2, *B*). Overall, tryptase levels were also increased in patients with MPCM with confluent compared with not confluent lesions (Table II).

In addition to patients with MPCM-small lesions, all patients with DCM exhibited significantly increased tryptase levels at initial presentation (Fig 2, *A*, and Table III). This also held true for patients who showed a decrease in their cutaneous involvement over time (Table III). Because improvement of DCM was regularly also associated with decreasing tryptase levels (Fig 2, *C*), we hypothesize that increased tryptase levels in these patients are mainly related to pronounced MC infiltration of the skin rather than to systemic involvement.⁴⁷⁻⁴⁹ Of 12 patients with DCM, 6 exhibited the rare subform of familial mastocytosis (Table III). Tryptase levels at first presentation were significantly lower in patients with familial compared with those with sporadic DCM. Confirming previous reports,^{50,51} we also found that tryptase levels in patients with familial DCM remained stable over 7 to 11 years and that skin lesions did not regress (Fig 2, *C*). Of note, we defined DCM strictly as homogeneous

thickening of the skin typically associated with prominent dermographism and a slightly yellow skin color (Fig 1), whereas previous studies, as assumed from published patient pictures, might have included patients with atypical MPCM lesions as well.^{47,49} Together, our findings on DCM suggest considering familial mastocytosis or a newly developed germline *KIT* mutation in patients with DCM,⁵² especially when tryptase levels remain stable.

Within the group of patients with MPCM-large lesions, we noticed several patients who exhibited nodular or plaque-type skin lesions at initial presentation, which usually flattened over time, as depicted for 1 patient in Fig 4, *B* (upper panel). In contrast, most patients with MPCM-small lesion had rather flat macular lesions at first presentation. This discrepancy might explain the significant difference in disease onset between elevated and flat lesions (Table II) because patients with MPCM-large lesions also had a significantly earlier disease onset than those with MPCM-small lesions.

We next focused on disease duration (Fig 3) and improvement of cutaneous lesions (Fig 4), which is an important issue in counseling families with children with mastocytosis. We found that patients with MPCM-large lesions compared with those with MPCM-small lesions have a significantly shorter disease duration (Fig 3, *A* and *B*) and more frequently experience

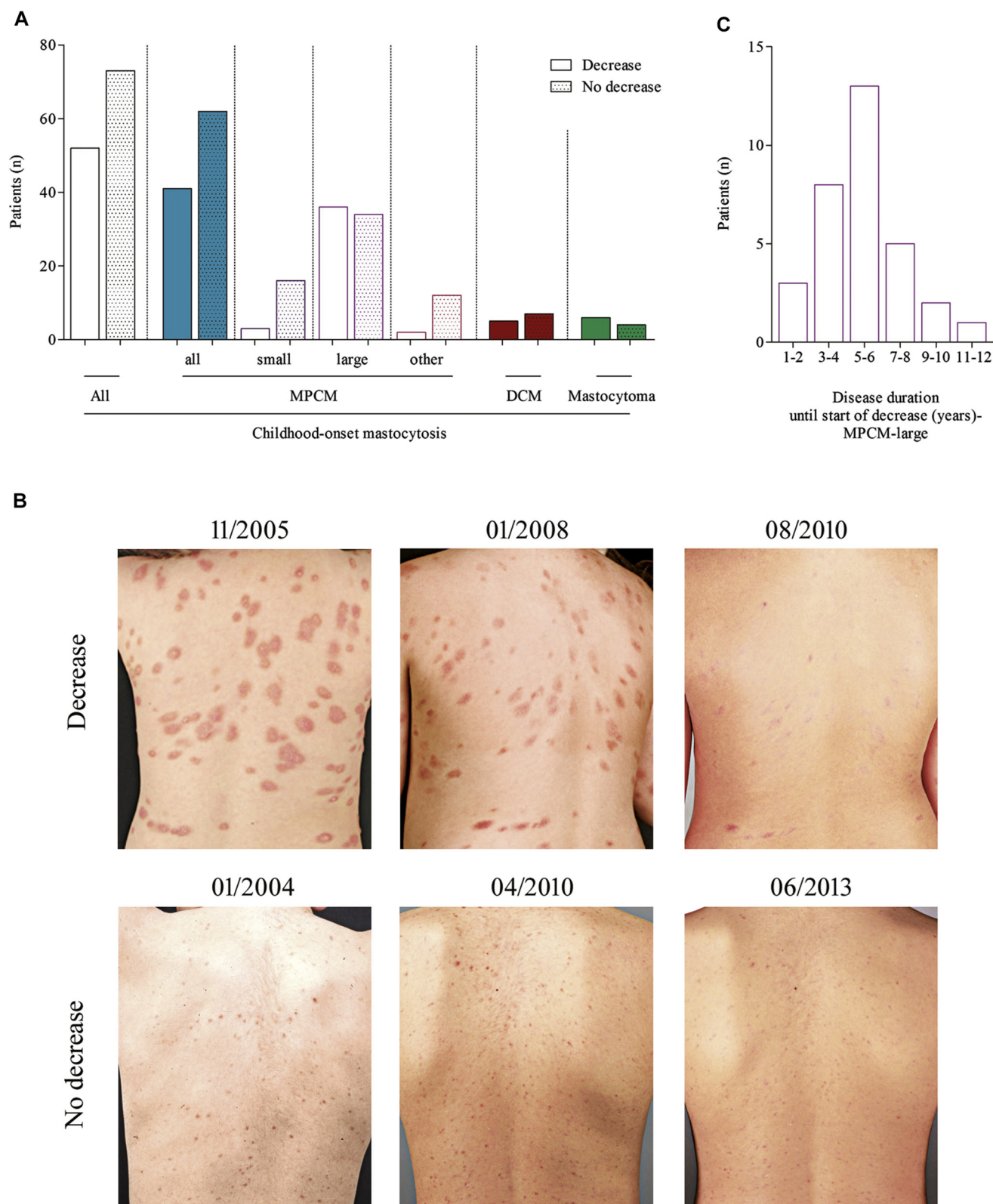


FIG 4. Spontaneous regression of skin lesions occurs more frequently in patients with MPCM-large lesions.

A, Number of patients with either a decrease (white bars) or no decrease (dotted bars) of skin lesions. **B**, Representative patient with MPCM-large lesions (upper panel) showing spontaneous improvement of skin lesions within 5 years (left, year 2005; middle, year 2008; right, year 2010) compared with a representative patient with MPCM-small lesions (lower panel) with no decrease in skin lesions over 9 years (left, year 2004; middle, year 2010; right, year 2013). **C**, Number of patients with MPCM-large lesions grouped according to disease duration from diagnosis until the start of decrease in skin lesions.

improvement of their skin lesions (Fig 4, A). Therefore it is likely that patients with MPCM-large lesions represent the subgroup of children with spontaneous regression of mastocytosis,²⁵⁻³⁰ whereas patients with MPCM-small lesions might represent those children with persistent mastocytosis until adulthood.³¹ Of note, MPCM-large lesions also started in most cases during infancy, whereas MPCM-small lesions often had disease onset after the age of 2 years (see Fig E2), which additionally suggests that MPCM-small lesions might, at least in some patients, represent an early form of adulthood-onset mastocytosis. However, the observation period of our patient population was too short to confirm this hypothesis. Further studies, especially prospective studies, are needed to validate this conclusion.

It might also be of importance for counseling of families that improvement of skin lesions in patients with MPCM-large lesions started after a disease duration of 5 to 6 years on average (Fig 4, C). To our knowledge, duration until the start of decrease in skin lesions has only been investigated in one other study.⁴⁵ Considering that patients with large MPCM lesions usually have onset of disease within the first 6 months of life and show a mean disease duration of 5.9 ± 4.8 years (Table II), it can be assumed that a decrease in their skin lesions will be visible until the age of 8 to 9 years.

The factors that determine the different morphology and course of disease in patients with childhood-onset mastocytosis are largely unknown. One could speculate that the different *KIT* mutations described in mastocytosis are associated with (or at least partially contribute to) specific variants of CM.^{16,18,20,22} As mentioned above, most adult patients express the mutation *KIT* D816V affecting exon 17 of the *KIT* gene, whereas pediatric patients frequently carry other mutations in exon 17, such as *KIT* D816F, D816Y, and D816I, or mutations in other exons, such as exons 8, 9, 10, and 11 (eg *KIT* S476I, ITD502-503, and K509I).^{15,16,18-21} In addition, there are pediatric patients who do not express any *KIT* mutation.^{15,16,21} Interestingly, Yang et al¹⁷ have demonstrated *in vitro* that mutations affecting either exon 17 or exons 8 and 9 display different signaling properties and whole-genome transcriptional profiles. Unfortunately, the studies exploring mutations in children did not describe the skin lesions in detail.^{16,18,20,21} In our study, investigating skin biopsy specimens from 28 patients for the presence of a *KIT* mutation in codon 816, we did not observe significant correlations between mutational status and subform of CM (see Fig E3). Future investigations should address this further and also screen for other *KIT* mutations outside of codon 816 in a larger group of patients with childhood-onset mastocytosis. Furthermore, we were only able to perform mutational analysis in skin biopsy specimens, which might lead to different results than analyzing bone marrow.

In the present study we demonstrate that patients with childhood-onset disease with large compared with small MPCM lesions show a more favorable outcome. This observation implies using the size of cutaneous lesions as a prognostic factor, which might in the long term serve to individualize counseling, follow-up, and treatment in patients with childhood-onset MPCM.

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Key messages

- Patients with childhood-onset MPCM present with heterogeneous skin lesions.
- Patients with childhood-onset disease with large maculopapular cutaneous lesions compared with small lesions have lower tryptase levels, shorter disease duration, earlier disease onset, and more frequent spontaneous regression of skin lesions.
- The size of maculopapular cutaneous lesions could serve as a prognostic parameter in patients with childhood-onset mastocytosis.

REFERENCES

1. Valent P. Mastocytosis: a paradigmatic example of a rare disease with complex biology and pathology. *Am J Cancer Res* 2013;3:159-72.
2. Horny HP, Akin C, Metcalfe DD, Escribano L, Bennett JM, Valent P, et al. Mastocytosis (mast cell disease). In: Swerdlow P, Campo P, Harris P, editors. World Health Organization (WHO) classification of tumours pathology and genetics of tumours of the haematopoietic and lymphoid tissues, 2. Lyon (France): IARC Press; 2008. pp. 54-63.
3. Akin C, Valent P. Diagnostic criteria and classification of mastocytosis in 2014. *Immunol Allergy Clin North Am* 2014;34:207-18.
4. Valent P, Akin C, Escribano L, Fodinger M, Hartmann K, Brockow K, et al. Standards and standardization in mastocytosis: consensus statements on diagnostics, treatment recommendations and response criteria. *Eur J Clin Invest* 2007;37:435-53.
5. Valent P, Horny HP, Li CY, Longley JB, Metcalfe DD, Parwaresch RM, et al. Mastocytosis. In: Jaffe RM, Harris RM, Stein RM, Vardiman RM, editors. World Health Organization (WHO) classification of tumours pathology and genetics of tumours of the haematopoietic and lymphoid tissues, 1. Lyon (France): IARC Press; 2001. pp. 291-302.
6. Valent P, Horny HP, Escribano L, Longley BJ, Li CY, Schwartz LB, et al. Diagnostic criteria and classification of mastocytosis: a consensus proposal. *Leuk Res* 2001;25:603-25.
7. Meni C, Bruneau J, Georgin-Lavialle S, Le Sache de Peuffelhous L, Damaj G, Hadj-Rabia S, et al. Paediatric mastocytosis: a systematic review of 1747 cases. *Br J Dermatol* 2015;172:642-51.
8. Frieri M, Quershi M. Pediatric mastocytosis: a review of the literature. *Pediatr Allergy Immunol Pulmonol* 2013;26:175-80.
9. Torrelo A, Alvarez-Twose I, Escribano L. Childhood mastocytosis. *Curr Opin Pediatr* 2012;24:480-6.
10. Lange M, Nedoszytko B, Górska A, Zawrocki A, Sobjanek M, Kozłowski D. Mastocytosis in children and adults: clinical disease heterogeneity. *Arch Med Sci* 2012;8:533-41.
11. Castells M, Metcalfe DD, Escribano L. Diagnosis and treatment of cutaneous mastocytosis in children: practical recommendations. *Am J Clin Dermatol* 2011;12:259-70.
12. Carter MC, Metcalfe DD. Paediatric mastocytosis. *Arch Dis Child* 2002;86:315-9.
13. Hartmann K, Metcalfe DD. Pediatric mastocytosis. *Hematol Oncol Clin North Am* 2000;14:625-40.
14. Kettelhut BV, Metcalfe DD. Pediatric mastocytosis. *J Invest Dermatol* 1991;96:15S-65S.
15. Bibi S, Langenfeld F, Jeanningros S, Brenet F, Soucie E, Hermine O, et al. Molecular defects in mastocytosis: KIT and beyond KIT. *Immunol Allergy Clin North Am* 2014;34:239-62.
16. Bodemer C, Hermine O, Palmerini F, Yang Y, Grandpeix-Guyodo C, Leventhal PS, et al. Pediatric mastocytosis is a clonal disease associated with D816V and other activating c-KIT mutations. *J Invest Dermatol* 2010;130:804-15.
17. Yang Y, Létard S, Borge L, Chaix A, Hanssens K, Lopez S, et al. Pediatric mastocytosis-associated KIT extracellular domain mutations exhibit different functional and signaling properties compared with KIT-phosphotransferase domain mutations. *Blood* 2010;116:1114-23.
18. Lantermier F, Cohen-Akenine A, Palmerini F, Feger F, Yang Y, Zermati Y, et al. Phenotypic and genotypic characteristics of mastocytosis according to the age of onset. *PLoS One* 2008;3:e1906.

19. Orfao A, Garcia-Montero AC, Sanchez L, Escribano L. Recent advances in the understanding of mastocytosis: the role of KIT mutations. *Br J Haematol* 2007; 138:12-30.
20. Yanagihori H, Oyama N, Nakamura K, Kaneko F. c-kit Mutations in patients with childhood-onset mastocytosis and genotype-phenotype correlation. *J Mol Diagn* 2005;7:252-7.
21. Sotlar K, Escribano L, Landt O, Mohrle S, Herrero S, Torrelo A, et al. One-step detection of c-kit point mutations using peptide nucleic acid-mediated polymerase chain reaction clamping and hybridization probes. *Am J Pathol* 2003;162:737-46.
22. Nagata H, Worobec AS, Oh CK, Chowdhury BA, Tannenbaum S, Suzuki Y, et al. Identification of a point mutation in the catalytic domain of the protooncogene c-kit in peripheral blood mononuclear cells of patients who have mastocytosis with an associated hematologic disorder. *Proc Natl Acad Sci U S A* 1995;92: 10560-4.
23. Brockow K, Jofer C, Behrendt H, Ring J. Anaphylaxis in patients with mastocytosis: a study on history, clinical features and risk factors in 120 patients. *Allergy* 2008;63:226-32.
24. Carter MC, Metcalfe DD, Clark AS, Wayne AS, Maric I. Abnormal bone marrow histopathology in paediatric mastocytosis. *Br J Haematol* 2015;168:865-73.
25. Lange M, Niedoszytko M, Renke J, Glen J, Nedoszytko B. Clinical aspects of paediatric mastocytosis: a review of 101 cases. *J Eur Acad Dermatol Venereol* 2013;27:97-102.
26. Ben-Amitai D, Metzker A, Cohen HA. Pediatric cutaneous mastocytosis: a review of 180 patients. *Isr Med Assoc J* 2005;7:320-2.
27. Kiszewski AE, Duran-Mckinster C, Orozco-Covarrubias L, Gutierrez-Castrellon P, Ruiz-Maldonado R. Cutaneous mastocytosis in children: a clinical analysis of 71 cases. *J Eur Acad Dermatol Venereol* 2004;18:285-90.
28. Middelkamp Hup MA, Heide R, Tank B, Mulder PG, Oranje AP. Comparison of mastocytosis with onset in children and adults. *J Eur Acad Dermatol Venereol* 2002;16:115-20.
29. Hannaford R, Rogers M. Presentation of cutaneous mastocytosis in 173 children. *Australas J Dermatol* 2001;42:15-21.
30. Azaña JM, Torrelo A, Mediero IG, Zambrano A. Urticaria pigmentosa: a review of 67 pediatric cases. *Pediatr Dermatol* 1994;11:102-6.
31. Berezowska S, Flaig MJ, Rueff F, Walz C, Haferlach T, Krokowski M, et al. Adult-onset mastocytosis in the skin is highly suggestive of systemic mastocytosis. *Mod Pathol* 2014;27:19-29.
32. Akin C, Metcalfe DD. Systemic mastocytosis. *Annu Rev Med* 2004;55:419-32.
33. Brockow K, Akin C, Huber M, Metcalfe DD. Assessment of the extent of cutaneous involvement in children and adults with mastocytosis: relationship to symptomatology, tryptase levels, and bone marrow pathology. *J Am Acad Dermatol* 2003;48:508-16.
34. Hartmann K, Henz BM. Cutaneous mastocytosis—clinical heterogeneity. *Int Arch Allergy Immunol* 2002;127:143-6.
35. Hartmann K, Henz BM. Mastocytosis: recent advances in defining the disease. *Br J Dermatol* 2001;144:682-95.
36. Wolff K, Komar M, Petzelbauer P. Clinical and histopathological aspects of cutaneous mastocytosis. *Leuk Res* 2001;25:519-28.
37. Soter NA. Mastocytosis and the skin. *Hematol Oncol Clin North Am* 2000;14:537-55.
38. Sperr WR, Jordan JH, Fiegl M, Escribano L, Bellas C, Dirnhofer S, et al. Serum tryptase levels in patients with mastocytosis: correlation with mast cell burden and implication for defining the category of disease. *Int Arch Allergy Immunol* 2002;128:136-41.
39. Erben P, Schwaab J, Metzgeroth G, Horny HP, Jawhar M, Sotlar K, et al. The KIT D816V expressed allele burden for diagnosis and disease monitoring of systemic mastocytosis. *Ann Hematol* 2014;93:81-8.
40. Hoermann G, Gleixner KV, Dinu GE, Kundi M, Greiner G, Wimazal F, et al. The KIT D816V allele burden predicts survival in patients with mastocytosis and correlates with the WHO type of the disease. *Allergy* 2014; 69:810-3.
41. Kristensen T, Broesby-Olsen S, Vestergaard H, Bindslev-Jensen C, Moller MB. Serum tryptase correlates with the KIT D816V mutation burden in adults with indolent systemic mastocytosis. *Eur J Haematol* 2013;91:106-11.
42. Kristensen T, Vestergaard H, Moller MB. Improved detection of the KIT D816V mutation in patients with systemic mastocytosis using a quantitative and highly sensitive real-time qPCR assay. *J Mol Diagn* 2011;13:180-8.
43. Chatzipetrou A, Koulias C, Zeliou CG, Potika M, Chliva K, Makris M. PD34—childhood mastocytosis: serum baseline total tryptase levels and extent of cutaneous disease as predictors of mast cell mediator release symptoms. *Clin Transl Allergy* 2014;28:34.
44. Alvarez-Twose I, Vano-Galvan S, Sanchez-Munoz L, Morgado JM, Matito A, Torrelo A, et al. Increased serum baseline tryptase levels and extensive skin involvement are predictors for the severity of mast cell activation episodes in children with mastocytosis. *Allergy* 2012;67:813-21.
45. Caplan RM. The natural course of urticaria pigmentosa. Analysis and follow-up of 112 cases. *Arch Dermatol* 1963;87:146-57.
46. Schwartz LB, Sakai K, Bradford TR, Ren S, Zweiman B, Worobec AS, et al. The alpha form of human tryptase is the predominant type present in blood at baseline in normal subjects and is elevated in those with systemic mastocytosis. *J Clin Invest* 1995;96:2702-10.
47. Lange M, Niedoszytko M, Nedoszytko B, Lata J, Trzeciak M, Biernat W. Diffuse cutaneous mastocytosis: analysis of 10 cases and a brief review of the literature. *J Eur Acad Dermatol Venereol* 2012;26:1565-71.
48. Koga H, Kokubo T, Akaishi M, Iida K, Korematsu S. Neonatal onset diffuse cutaneous mastocytosis: a case report and review of the literature. *Pediatr Dermatol* 2011;28:542-6.
49. Heide R, Zuidema E, Beishuizen A, Den Hollander JC, Van Gysel D, Seyger MM, et al. Clinical aspects of diffuse cutaneous mastocytosis in children: two variants. *Dermatology* 2009;219:309-15.
50. Hartmann K, Wardelmann E, Ma Y, Merkelbach-Bruse S, Preussner LM, Woolery C, et al. Novel germline mutation of KIT associated with familial gastrointestinal stromal tumors and mastocytosis. *Gastroenterology* 2005;129: 1042-6.
51. Tang X, Boxer M, Drummond A, Ogston P, Hodgins M, Burden AD. A germline mutation in KIT in familial diffuse cutaneous mastocytosis. *J Med Genet* 2004; 41:e88.
52. Akin C, Fumo G, Yavuz AS, Lipsky PE, Neckers L, Metcalfe DD. A novel form of mastocytosis associated with a transmembrane c-kit mutation and response to imatinib. *Blood* 2004;103:3222-5.

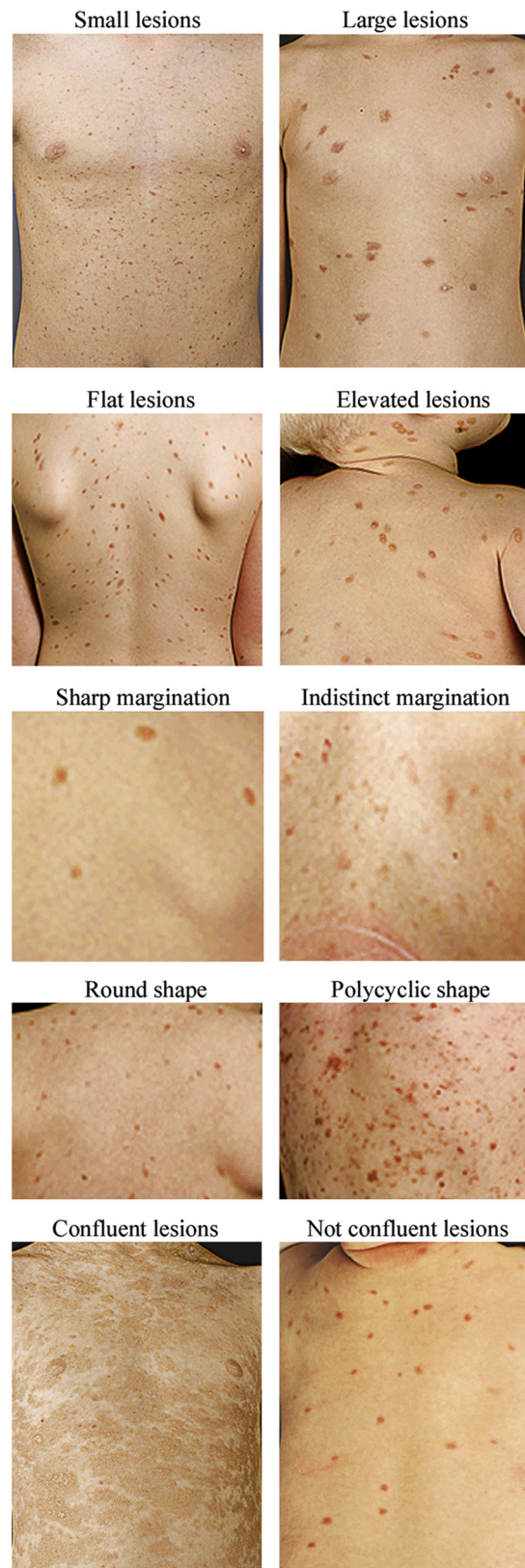


FIG E1. Representative pictures of dermatologic terms used within the text.

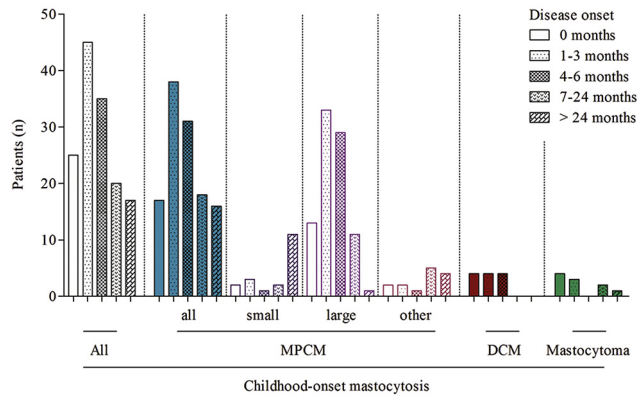


FIG E2. Number of patients grouped according to disease onset.

No.	Patient characteristics					Mutational and histological characteristics in skin biopsy							
	Gender	Disease onset (months)	Disease duration (years)	Tryptase (µg/l)	Subform of CM and MPCM size	<i>KIT</i> mutational status	MC density	MC localization	MC aggregates	MC infiltration pattern	MC morphology	Spindle-shaped MC (%)	CD25+ MC (%)
119	f	168	9	16,10	small	wt	1	pb, pv	no	a	mostly round	20	10
7	f	2	42	86,30	small	wt	1	pv, (pb, rb)	no	a	mostly spindle-shaped	70	10
118	f	174	2	5,00	small	D816V	2	pb, pv	no	b	mostly spindle-shaped	90	0
63	f	168	13	7,00	small	D816V	2	pb, pv, pa	no	a	mostly spindle-shaped	60	0
75	m	4	3	n.d.	large	wt	2	pb, pv, pa	no	a	mostly round	30	10
123	f	6	9	n.d.	large	wt	3	pb, pv, pa	yes	b	mostly round	10	70
109	m	1	2	n.d.	large	wt	1	pb, rb	no	c	mostly round	10	n.a.
64	m	2	18	n.d.	large	wt	1	pv, (rb, pb)	no	a	mostly spindle-shaped	90	20
110	f	1	3	1,44	large	n.a.	3	pb, rb	yes	d	mostly round	5	10
19	m	0	8	3,03	large	wt	2	pb, (pv)	yes	b	round	10	80
25	f	0	2	3,14	large	wt	2	pb, pv, pa	no	a	mostly spindle-shaped	80	10
58	m	17	7	3,50	large	wt	2	pb, pv, pa	no	a	mostly spindle-shaped	50	10
24	f	3	6	4,50	large	wt	3	pb, (pv)	yes	b	mostly round	10	60
69	m	6	1	5,98	large	wt	2	pv, (pb)	no	a	mostly spindle-shaped	60	30
92	f	0	3	7,63	large	wt	3	pb, pv, pa	yes	b	round	0	90
16	f	12	12	7,91	large	n.a.	3	rb, pb, pv	no	c	mostly spindle-shaped	70	30
93	f	14	7	10,40	large	wt	3	pb, pv, pa	no	a	mostly spindle-shaped	70	30
136	m	0	2	16,60	large	wt	3	pb, rb	yes	d	round	0	0
108	f	0	3	20,30	large	wt	3	pb, rb	yes	d	round	0	30
53	f	5	8	n.d.	large	D816V	1	rb, pb	no	b	mostly spindle-shaped	80	0
43	f	3	6	3,30	large	D816V	2	pb, pv, (rb)	no	b>a	mostly spindle-shaped	70	0
79	m	1	10	3,94	large	D816V	2	pb, pv	yes	b	mostly round	20	0
91	f	5	22	4,03	large	D816V	3	pb, pv, pa	yes	b	round	0	0
82	m	6	2	5,00	large	D816V	1	pv	no	c	mostly round	20	0
30	m	2	15	8,41	large	D816V	2	pv, (pb, rb)	no	a	mostly round	30	10
26	m	1	9	n.d.	large	D816Y	1	rb, pv	no	a	mostly round	40	0
96	m	24	10	32,20	other	D816V	3	rb, pb, pv, pa	yes	d	round	0	100
10	f	5	4	63,00	DCM	wt	3	pb, (rb)	yes	b>d	round	0	70
78	m	4	7	100,00	DCM	wt	3	pb, rb	yes	d	round	0	30
116	m	6	5	177,00	DCM	wt	3	pb, pv	yes	b	round	0	90

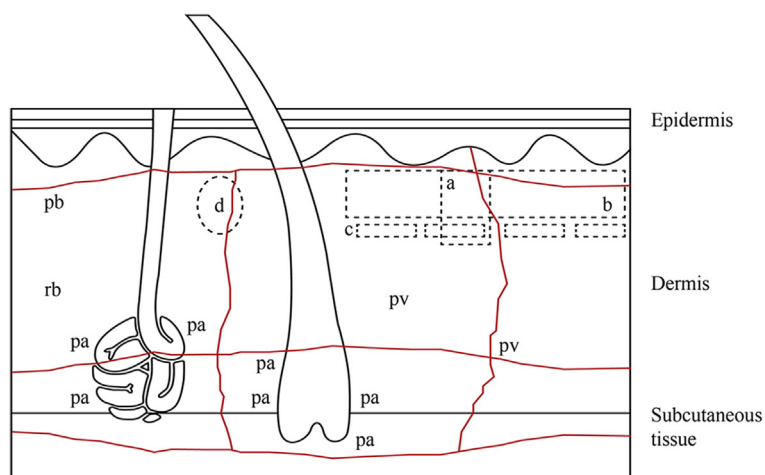


FIG E3. Mutational and histologic characteristics in a subset of patients participating in this study. *large*, MPCM-large lesions; *n.a.*, not applicable; *n.d.*, not detected; *other*, MPCM-other lesions; *small*, MPCM-small lesion; *wt*, wild type. The *blue-red* color scale illustrates hierarchic comparisons: *blue*, minimum; *red*, maximum. MC density is as follows: 1, low; 2, middle; 3, high. MC localization is as follows: *pa*, periadnexal (around hair follicles/sweat glands); *pb*, papillary body; *pv*, perivascular; *rb*, reticular body. MC infiltration pattern is as follows: *a*, perivascular in the papillary body and upper reticular dermis; *b*, sheet-like within the papillary body and upper reticular dermis; *c*, interstitial; *d*, nodular.