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Original Article

Systemic mastocytosis: A cohort study on clinical characteristics of 136 patients in a large tertiary centre

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ABSTRACT

Background: Systemic mastocytosis (SM) is a rare heterogeneous disease which is characterized by the aberrant proliferation of mast cells. It can be divided into various subtypes with different phenotypes and prognoses. Here, we report on the clinical characteristics of 136 SM patients.
Methods: A retrospective cohort study was conducted from January 2009 to September 2014 in a large tertiary centre in The Netherlands. We included all patients who fulfilled WHO criteria for SM. Data were collected from electronic patient files.
Results: A total of 124 patients had indolent SM (ISM) (91.2%), 7 had aggressive SM (ASM) (5.1%) and 5 had SM with associated hematological non-mast cell lineage disease (SM-AHNMD) (3.7%). There was no progression from ISM to advanced SM subtypes, but 1 patient with ASM developed chronic myelocytic leukemia 2 years after diagnosis. The average time to diagnosis for the whole population was 8.1 years (range, 0–49 years). The most frequent triggers for work-up—skin involvement, anaphylaxis and osteoporosis—were characterized by an interval to diagnosis of 10.9, 2.9 and 7.5 years, respectively. A total of 32 patients (23.5%) had a serum tryptase levels below the cutoff value of 20 ng/mL at the time of diagnosis, but these patients did not have significant differences in clinical phenotype.
Conclusions: SM comprises a wide spectrum of signs and symptoms and its often atypical presentation can delay the establishment of the diagnosis substantially. Skin involvement, anaphylaxis and unexplained osteoporosis should trigger analysis for mastocytosis. A normal serum tryptase does not exclude the diagnosis of SM.

1. Introduction

Mastocytosis is a rare systemic disease which is characterized by uncontrolled proliferation of aberrant mast cells [1]. According to the definition of the World Health Organization (WHO) it is a myeloproliferative disease with different subtypes [2]. In systemic mastocytosis (SM), at least one extracutaneous organ is affected. Systemic mastocytosis is divided in various subtypes (Table 1). Most patients have indolent SM (ISM), which generally has a mild course and does not affect overall survival. It is increasingly recognized that ISM patients with or without skin lesions (ISMs+ or ISMs−, respectively) have clinically distinct phenotypes [3]. Smouldering SM (SSM) is a relatively new subtype of ISM and is defined by the presence of organ involvement without organ dysfunction. In SM with associated hematological non-mast cell lineage disease (SM-AHNMD), the prognosis is determined by the associated condition. Furthermore, aggressive SM (ASM) is characterized by organ dysfunction due to infiltration of mast cells. This subtype often needs more intensive treatment with cytoreductive therapy.

The prevalence of SM in The Netherlands is approximately 13 in 100.000 residents, which appears to be in accordance with other European countries [4]. In 80%–90% of patients, the D816V mutation is found in the gene encoding for c-KIT, a tyrosine kinase that functions as stem cell factor (SCF) receptor. A small proportion of patients has other mutations in c-KIT. This mutation leads to uncontrolled proliferation and inhibition of apoptosis through continuous stimulation of c-KIT, even in the absence of SCF [5]. However, not every SM patient has the D816V mutation, and conversely, not all patients with the D816V mutation express the same phenotype. Therefore, other unknown factors also have to play a role in the pathogenesis of SM [6].

SM is known to cause a wide diversity of symptoms. These can vary from the typical urticaria pigmentosa or flushing and itching, to less specific symptoms like osteoporosis, diarrhea or unexplained syncope. Most symptoms are caused by high levels of mast cell mediators, mainly histamine and proinflammatory cytokines [7]. Due to this heterogeneous presentation, diagnosing this disease, particularly ISM, can be a true challenge which requires high clinical suspicion. That a delay in

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Table 1
Diagnostic criteria for systemic mastocytosis [2].

Systemic mastocytosis			
Presence of 1 major and 1 minor criterium or 3 minor criteria. Major criterium: • Multifocal, dense MC infiltrate (with >15 MC per infiltrate) in bone marrow and/or extracutaneous organ. Minor criteria: • Presence of D816V KIT mutation in bone marrow, peripheral blood or extracutaneous tissue. • Serum tryptase >20 μg/mL. • Expression of CD117 + either CD2 or CD25 in MC in bone marrow. • >25% atypical or spindle-shaped MC.			
ISM	SSM	SM-AHNMD	ASM
Fulfills criteria for SM, without the presence of B- or C-findings ISMs − ISM without skin lesions ISMs + ISM with skin lesions	Fulfills criteria for SM + > 2 B-findings: • Hepato- or splenomegaly without organ dysfuntion. • Lymphadenopathy. • >30% MC infiltration in bone marrow. • Serum tryptase >200 ng/mL. Signs of dysplasia or myeloproliferative disease without fulfilling criteria for SM-AHNMD.	Fulfills criteria for SM + a non-mast cell lineage clonal hematological disease (myelodysplastic syndrome, acute myeloid leukemia, non-Hodgkin lymphoma or myeloproliferative neoplasm).	Fulfills criteria for SM + >1 'C-findings': • Anemia, neutropenia of thrombocytopenia* • Hepatomegaly, ascites, hepatic dysfunction and/or portal hypertension. • Malabsorption. • Bone involvement with pathological fracture and/or osteolytical bone lesions. Splenomegaly with hypersplenism.

MC, mast cells; SM, systemic mastocytosis; ISM, indolent systemic mastocytosis; SSM, smouldering systemic mastocytosis; ASM, aggressive systemic mastocytosis; SM-AHNMD, systemic mastocytosis with associated hematological non-mast cell lineage disease.
* Anemia Hb <10 g/dL, neutropenia absolute neutrophil count $<50 \times 10^6$, thrombopenia $<1500 \times 10^6$.

the diagnostic process can lead to several undesired consequences like organ dysfunction, life-threatening anaphylaxis or severe osteoporosis needs no further explanation [8,9].

The objective of this study is to describe the clinical characteristics of all patients with SM who were referred to the Erasmus University Medical Centre in the last 5 years. Hereby, we hope to create a better understanding of the way patients present to us and hopefully improve the diagnostic process.

2. Methods

2.1. Patient selection and follow-up

We selected all patients who visited the Erasmus MC University Medical Centre from January 2009 to September 2014 and fulfilled the WHO criteria for systemic mastocytosis [2]. Recently, the NFU (Dutch federation of academic medical centres) classified the Erasmus MC Mastocytosis Centre as a centre of excellence. Included patients could have visited either the outpatient clinic of clinical immunology, hematology or allergology. Therefore, no uniform diagnostic work-up was performed. Also, guidelines on the work-up of SM have changed in the studied period. However, general laboratory tests including serum tryptase and bone marrow examination was performed in most patients. Patients were routinely seen once yearly for follow-up purposes in the outpatient clinic. Yearly, data on clinical symptoms, laboratory markers (including serum tryptase) and an abdominal ultrasound were performed to screen for progression. Once in every 2 years, bone densitometry was performed for follow-up on osteoporosis. By retrospectively studying the patient files, we collected a wide array of data: the symptom that triggered referral and analysis, the time of diagnosis, other mastocytosis-related symptoms, various laboratory results and organ involvement. Most patients were followed-up in our centre. For the patients who were not followed-up in our centre, we only retrieved information on their visits in our centre and on their survival until September 2014.

2.2. Definitions

For the different subtypes of SM, we used the WHO classification [2]. For skin involvement, both urticaria pigmentosa and telangiectasia macularis eruptiva perstans were accounted for. Neuropsychiatric symptoms were defined as every psychiatric diagnosis in the patient's medical history or the current use of psychiatric medication. Cytopenia involved anemia (Hb <10 g/dL), leukopenia ($<400 \times 10^6$) and/or thrombocytopenia ($<1500 \times 10^6$). Osteoporosis was classified according to *T*-scores: osteoporosis = *T* score ≤−2.5; osteopenia = *T* score −1 to −2.5; osteosclerosis = T-score >+2.5. A pathological fracture was defined as a spontaneous fracture directly linked to SM. Osteoporotic vertebral fractures were not included in this definition.

2.3. Statistical analysis

We used IBM Statistics SPSS 21 for all analyses. Frequencies, percentages with range or standard deviation were calculated for all variables. The subtypes of SM were compared with a one-way ANOVA test for continuous variables and with a chi-square test for dichotomous variables. Correlation coefficients were calculated with Spearman's rho.

2.4. Mutational analysis

In brief, 400 μL of EDTA-preserved bone marrow was used to isolate DNA with MagNApure (Roche Molecular Systems, Mannheim, Germany) according to the manufacturer's instructions. Subsequently, a standard solution of 50 ng/μL was prepared using the Nandrop (Shimadzu Corporation, Kyoto, Japan) of which 3 μL was used for each analysis. Detection of D816V KIT mutation was performed by two independent methods in all samples: the LightCycler System (Roche Molecular Systems, Mannheim, Germany) for the primary result and the TaqMan System (Fisher Scientific, Amsterdam, The Netherlands) for confirmation. In the LightCycler assay, the amplification rate of mutated DNA over wild-type DNA was improved by adding Locked Nucleic Acid (LNA, Tib Molbiol, Berlin, Germany). The mutation was detected by melting point analysis. In the TaqMan assay, a mismatched positive primer with a much higher affinity for the mutant DNA compared to wild-type DNA was used. A cycle time (CT) value <40 was considered as positive for the presence of D816V. In both assays, the detection limit is 0.1% mutated copies. Two negative controls (a blank without DNA and one with wild-type DNA) and two positive controls (one with diluted plasmid DNA of the mutation to assess the detection limit and one with the D816V mutation) were used. The wild-type and