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Inclusion body myositis with granuloma formation in muscle tissue

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Abstract

Inclusion body myositis is a form of inflammatory myopathy. We identified 4 cases of inclusion body myositis showing granuloma formation in muscle tissue and aimed to assess the features of this atypical form of inclusion body myositis. We retrospectively reviewed consecutive patients who satisfied European Neuromuscular Centre IBM Research Diagnostic Criteria 2011. Then, we assessed clinical profiles and pathological findings in patients with inclusion body myositis with granuloma and compared these findings with those of typical inclusion body myositis without granuloma. We identified 15 patients with inclusion body myositis. Four patients showed granuloma formation in muscle tissue in addition to typical pathological features of inclusion body myositis. Granulomas comprised a mixture of inflammatory cells, such as macrophages, epithelioid histiocytic cells, and lymphocytes. One patient was found to have mediastinal granulomatous lymphadenopathy; however, the evidence in other patients was insufficient for a diagnosis of systemic sarcoidosis. There were no significant differences between groups with and without granuloma regarding clinical manifestations, laboratory findings, response to immunomodulating therapies, or myopathological profiles. We established a new form of inclusion body myositis showing granuloma formation in muscle tissue. Inclusion body myositis and granuloma formation could have identical pathomechanisms concerning dysregulation of autophagy.

Keywords: Inclusion body myositis; Granuloma; Rimmed vacuole; Autophagy

1. Introduction

Inclusion body myositis (IBM) is a type of inflammatory myopathy characterized by T lymphocyte infiltration around muscle fibers and the presence of rimmed vacuoles [1]. In comparison to other types of inflammatory myopathy, including polymyositis and dermatomyositis, patients with IBM demonstrate an unfavorable response to immunosuppressive therapy [1]. The pathomechanisms of IBM remain uncertain. Inflammatory processes and degenerative mechanisms are both responsible for the development of this muscular disorder.

Granulomas are pathological structures comprising giant cells, macrophage-like epithelioid cells, and lymphocytes. The most common condition associated with non-caseating granuloma formation in muscle tissues is sarcoidosis followed by foreign-body reactions and infectious conditions; however, granulomatous myositis (GM) is a type of inflammatory myopathy containing granulomas without any evidence of systemic sarcoidosis despite intensive clinical investigations [2].

Although there are several reported cases that presented features of both IBM and sarcoidosis [3–5], the relationship between IBM and granuloma formation remains to be clarified. In this study, we identified 4 patients with IBM showing granuloma formation in muscle tissues. Somewhat surprisingly, granulomas were confined to skeletal muscle in 3 patients. This study aimed to elucidate the features of IBM with granuloma by comparing these features with the clinical and histopathological features of patients with typical IBM.

2. Materials and methods

2.1. Patients

We retrospectively reviewed consecutive patients who underwent an open muscle biopsy in our hospital and were referred to our department for pathological diagnosis of muscle biopsies between 2003 and 2013. We selected IBM patients who satisfied the European Neuromuscular Centre (ENMC) IBM Research Diagnostic Criteria 2011 [6], and applied the

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classification as follows: clinicopathologically defined IBM, clinically defined IBM, and probable IBM.

Medical records were retrospectively reviewed for age at diagnosis, sex, sites of initial weakness, and duration from onset to diagnosis. The following laboratory features determined at the time of diagnosis were obtained: the levels of serum creatine kinase, lactate dehydrogenase, aldolase, and myoglobin; levels of plasma C-reactive protein; and erythrocyte sedimentation rate. In addition, we examined the presence of serum autoantibodies suggestive of other collagen disorders, hepatitis B antigens, and hepatitis C antibodies.

Special attention was paid to patients showing granulomas in muscle to exclude the possibility of systemic sarcoidosis. A clinical diagnosis of sarcoidosis could be made on the basis of clinical, laboratory, and radiological features in addition to evidence of non-caseating granulomas in tissues. Moreover, it is essential to exclude alternative diseases [7]. In terms of clinical presentation, pulmonary symptoms and abnormal chest radiographs including bilateral hilar lymphadenopathy are common; however, rarely patients may show only extrapulmonary manifestations [7]. Granulomatous myositis is a type of myositis characterized by granuloma formation in muscle tissue, variable response to immunosuppressive therapies, and no evidence of systemic sarcoidosis [2]. Although it is uncertain whether GM is a limited form of systemic sarcoidosis, not all patients with GM develop symptoms of sarcoidosis [8]. To avoid making a diagnosis of sarcoidosis, the patients with IBM showing granuloma underwent intensive examinations, which included ophthalmological examinations, chest radiographs, and whole body examinations. Other disorders which might cause granulomatous myositis, such as tuberculosis infection, lymphoma, intestinal inflammatory disease, myasthenia gravis, foreign-body reaction, cryofibrinogenemia, and primary biliary cirrhosis [2], were excluded carefully by performing radiological and clinical investigations.

Most patients received immunosuppressive therapy as follows: corticosteroids (prednisone and methylpredonisolone), immunosuppressive drugs, and intravenous immunoglobulin (IVIg). The results of these treatments were examined by retrospectively reviewing the medical records.

Written informed consent to participate in this study was obtained from the patients. The study protocol was approved by the medical ethics committee of Kanazawa University.

2.2. Pathological examinations

Muscle biopsies were performed in all patients. All samples were obtained by open biopsies and divided into several blocks. Some specimens were immediately frozen in isopentane cooled with liquid nitrogen and stored at −80 °C until the experiments. Serial 6-μm-thick cryostat sections were cut and stained with hematoxylin and eosin (HE), modified Gomori trichrome, NADH-tetrazolium, Congo Red, and cytochrome C oxidase (COX). The remaining samples were fixed with 10% buffered formalin. Pathological examinations were also performed on 5-μm-thick sections of paraffin embedded blocks using HE and Congo Red.

Selected formalin-fixed sections were immunostained with antibodies against phosphorylated neurofilaments (SMI-31; Covance, Emeryville, CA; 1:500), amyloid β protein (4G8; Covance; 1:5000), ubiquitin (Dako, Glöstrup, Denmark; 1:8000), macrophages (CD68; Dako; 1:100), T lymphocytes (CD3; Dako; 1:20), B lymphocytes (CD20; Dako; 1:50), helper T lymphocytes (CD4; Dako; 1:400), and cytotoxic T lymphocytes (CD8; Dako; 1:20) using appropriate antigen retrieval methods. Immunohistochemistry using antibodies against human leukocyte antigen-I (HLA-I; Dako; 1:200) and HLA-II (Dako, 1:100) was performed on frozen sections. The EnVision system (Dako) was used for these immunolabeling studies. Peroxidase labeling was visualized with diaminobenzidine as the chromogen.

Granulomas were defined as collections of inflammatory cells in which more than 60% of the inflammatory cells were macrophages or epithelioid histiocytic cells, as judged by HE or specific macrophage marker staining (CD68). The presence of Langerhans-type or histiocytic giant cells was unnecessary to be judged as granulomas [9]. We paid special attention to exclude necrotic regions to avoid including caseating granulomas [9].

2.3. Case classification

With reference to the pathological features of IBM (endomysial inflammatory infiltrates; rimmed vacuoles; protein accumulation; and upregulation of HLA class I around muscle fibers), we made a clinicopathological diagnosis of IBM [6]. Patients who demonstrated only these features were designated as the typical IBM without granuloma group. Patients showing granuloma formation in addition to typical IBM clinicopathological characteristics were categorized into the IBM with granuloma group.

2.4. Statistical analysis

Differences in age at disease onset and in the results of laboratory tests between the typical IBM without granuloma group and the IBM with granuloma group were assessed using a Mann–Whitney *U* test. Sex, diagnosis classification of IBM, initial weakness, and pathological findings were assessed using chi-square test and Fisher's exact probability test. Statistical significance was defined as $p < 0.05$. Statistical analyses were performed using SPSS version 22 (IBM, Armonk, NY).

3. Results

3.1. Clinical profiles

A total of 307 patients with myopathy were analyzed in our department during the study period. Fifteen patients satisfied the clinical diagnostic criteria of IBM [6]. Five patients were categorized as clinicopathologically defined IBM. Four and 6 patients were categorized as clinically defined IBM and probable IBM, respectively. Granuloma formation in muscle tissues was noted in 4 patients. In these patients, one patient met the criteria for clinicopathologically defined IBM, whereas 2 and 1 patients were categorized as clinically defined IBM and probable IBM, respectively.