Dysregulated mature IL-1beta production in familial Mediterranean fever	Migita K, Izumi Y, Fujikawa K, Agematsu K, Masumoto J, Jiuchi Y	Rheumatology	2014	国外
Clinical and laboratory features of fatal rapidly progressive interstitial lung disease associated with juvenile dermatomyositis	Kobayashi N, Takezaki S, Kobayashi I, Iwata N, Mori M, Nagai K, Nakano N1, Miyoshi M1, Kinjo N1, Murata T1, Masunaga K1, Umebayashi H1, Imagawa T1, Agematsu K1, Sato S1, Kuwana M1, Yamada M1, Takei S1, Yokota S1, Koike K1, Ariga T	Rheumatology	2014	国外
Clinical course in a patient with neutrophil-specific granule deficiency and rapid detection of neutrophil granules as a screening test	Shigemura T, Yamazaki T, Shiohara M, Kobayashi N, Naganuma K, Koike K, Agematsu K	Journal of clinical immunology	2014	国外
Dramatic Improvement in the Multifocal Positron Emission Tomography Findings of a Young Adult with Chronic Granulomatous Disease Following Allogeneic Hematopoietic Stem Cell Transplantation	Shigemura T, Nakazawa Y, Hirabayashi K, Kobayashi N, Sakashita K, Agematsu K	Journal of clinical immunology	2014	国外
IL-18 serum concentration is markedly elevated in typical familial Mediterranean fever with M694I mutation and can distinguish atypical type	Takashi Yamazaki, Tomonari Shigemura, Norimoto Kobayashi, Kimiko Honda, Masahide Yazakie, Junya Masumoto, Kiyoshi Migita, Kazunaga Agematsu	Modern Rheumatology	2015	国外
Familial Mediterranean fever with P369S/R408Q exon3 variant in pyrin presenting as symptoms of PFAPA	Keiko Yamagami, Tomoyuki Nakamura, Ryota Nakamura, Yusuke Hanioka, Kaori Seki, Hiroshi Chiba, Keiko Kobayashi, Kazunaga Agematsu	Modern Rheumatology	2015	国外
Successful treatment for West syndrome with severe combined immunodeficiency	Motobayashi M, Inaba Y, Fukuyama T, Kurata T, Niimi T, Saito S, Shiba N1, Nishimura T1, Shigemura T1, Nakazawa Y1, Kobayashi N1, Sakashita K1, Agematsu K4, Ichikawa M5, Koike K	Brain & development	2015	国外
わが国における家族性地中海熱の臨床像	上松 一永	「小児科」	2014年2月	国内
FAPA症候群の臨床像の検討	古本 雅宏、柴 直子、伯耆原 祥、重村 倫成、小林 法元、小池 健一、本多 貴実子、梅田 陽一、上松 一永	「日本小児科学会雑誌(0001-6543)」	2014年2月	国内

（注１）発表者氏名は、連名による発表の場合には、筆頭者を先頭にして全員を記載すること。
（注２）本様式はexcel形式にて作成し、甲が求める場合は別途電子データを納入すること。

様式第１９

学　会　等　発　表　実　績

委託業務題目「遺伝子変異に基づくFMFインフラマソーム病態解明と炎症制御に向けたトランスレーショナル研究」
機関名　長崎大学病院　第一内科
分担機関名　愛媛大学プロテオサイエンスセンター

１．学会等における口頭・ポスター発表

発表した成果（発表題目、口頭・ポスター発表の別）	発表者氏名	発表した場所（学会等名）	発表した時期	国内・外の別
Defect of suppression of inflammasome-independent interleukin-8 secretion from sw982 synovial sarcoma cells by familial mediterranean fever-derived pyrin mutations	Masumoto J	16th Biennial Meeting of the European Society for Immunodeficiencies	2014 October 30-November 1	国外

２．学会誌・雑誌等における論文掲載

掲載した論文（発表題目）	発表者氏名	発表した場所（学会誌・雑誌等名）	発表した時期	国内・外の別
Tocilizumab improved clinical symptoms of a patient with systemic tophaceous gout who had symmetric polyarthritis and fever: An alternative treatment by blockade of interleukin-6 signaling	Mokuda S, Kanno M, Takasugi K, Okumura C, Ito Y, Masumoto J	SAGE Open Medical Case Reports	2014	国外
Defect of suppression of inflammasome-independent interleukin-8 secretion from SW982 synovial sarcoma cells by familial Mediterranean fever-derived pyrin mutations	Sugiyama R, Agematsu K, Migita K, Nakayama J, Mokuda S, Ogura F, Haraikawa K, Okumura C, Suehiro S, Morikawa S, Ito Y, Masumoto J	Mol Biol Rep	2014	国外
Epstein-Barr virus-related MTX-LPD in rheumatoid arthritis patients exhibits a viral pattern of the CD64 and CD35 expression on neutrophils: Three case reports	Mokuda S, Miyazaki T, Saeki Y, Masumoto J, Kanno M, Takasugi K	Mod Rheumatol	2014	国外
Microvasculature of carotid atheromatous plaques: hemorrhagic plaques have dense microvessels with fenestrations to the arterial lumen	Kurata M, Nose M, Shimazu Y, Aoba T, Kohada Y, Yorioka S, Suehiro S, Fukuoka E, Matsumoto S, Watanabe H, Kumon Y, Okura T, Higaki J, Masumoto J	J Stroke Cerebrovasc Dis	2014	国外
Familial Mediterranean fever: genotype-phenotype correlations in Japanese patients	Migita K, Agematsu K, Yazaki M, Nonaka F, Nakamura A, Toma T, Kishida D, Uehara R, Nakamura Y, Jiuchi Y, Masumoto J, Furukawa H, Ida H, Terai C, Nakashima Y, Kawakami A, Nakamura T, Eguchi K, Yasunami M, Yachie A	Medicine (Baltimore)	2014	国外

Serum amyloid A induces NLRP-3-mediated IL-1β secretion in neutrophils	Migita K, Izumi Y, Jiuchi Y, Kozuru H, Kawahara C, Nakamura M, Nakamura T, Agematsu K, Masumoto J, Yasunami M, Kawakami A, Eguchi K	PLoS One	2014	国外
Dysregulated mature IL-1β production in familial Mediterranean fever	Migita K, Izumi Y, Fujikawa K, Agematsu K, Masumoto J, Jiuchi Y, Kozuru H, Nonaka F, Shimizu T, Nakamura T, Iwanaga N, Furukawa H, Yasunami M, Kawakami A, Eguchi K	Rheumatology (Oxford)	2014	国外
K.IL-18 serum concentration is markedly elevated in typical familial Mediterranean fever with M694I mutation and can distinguish it from atypical type	Yamazaki T, Shigemura T, Kobayashi N, Honda K, Yazaki M, Masumoto J, Migita K,Agematsu	Mod Rheumatol	2014	国外
IL-1 as a target in inflammation	Ito Y, Kaneko N, Iwasaki T, Morikawa S, Masumoto J	Endocr Metab Immune Disord Drug Targets	2014	国外

（注１）発表者氏名は、連名による発表の場合には、筆頭者を先頭にして全員を記載すること。
（注２）本様式はexcel形式にて作成し、甲が求める場合は別途電子データを納入すること。

Ⅳ. 研究成果の刊行物・別冊

Clinical & Experimental Immunology
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Increased prevalence of *MEFV* exon 10 variants in Japanese patients with adult-onset Still's disease

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Summary

Autoinflammatory diseases include a large spectrum of monogenic diseases, e.g. familial Mediterranean fever (FMF), as well as complex genetic trait diseases, e.g. adult-onset Still's disease (AOSD). In populations where FMF is common, an increased *MEFV* mutation rate is found in patients with rheumatic diseases. The aim of this study was to examine *MEFV* mutations in Japanese patients with AOSD. Genomic DNA was isolated from 49 AOSD patients and 105 healthy controls, and exons 1, 2, 3 and 10 of the *MEFV* gene genotyped by direct sequencing. *MEFV* mutation frequencies in AOSD patients were compared with controls. We found no significant difference in overall allele frequencies of *MEFV* variants between AOSD patients and controls. However, *MEFV* exon 10 variants (M694I and G632S) were significantly higher in AOSD patients than controls (6·1 *versus* 0%). In addition, there was no significant difference between *MEFV* variant carriers and non-carriers with clinical manifestations, but the monocyclic clinical course of the AOSD disease phenotype was observed less frequently in patients without *MEFV* variants. AOSD patients had significantly higher frequencies of *MEFV* exon 10 mutations, suggesting that low-frequency variants of *MEFV* gene may be one of the susceptibility factors of AOSD.

Keywords: adult-onset Still's disease, autoinflammatory disease, *MEFV*

Introduction

Adult-onset Still's disease (AOSD) is a systemic inflammatory disease characterized by fever, skin rash, and joint pain [1]. The pathophysiology of AOSD has remained unclear; however, it is presumed to be an autoinflammatory disease due to the absence of autoantibodies and autoantigen-specific T cells [2]. Patients with autoinflammatory diseases, including classical hereditary periodic syndromes, share clinical features (e.g. spiking fever, skin rash, and arthritis) with the major symptoms of AOSD [3]. Familial Mediterranean fever (FMF) is a common autoinflammatory disease, characterized by recurrent inflammatory attacks of fever, skin rash, synovitis, and serositis [4]. FMF is thought to be caused by gain-of-function mutations in the *MEFV* gene [5]. Although AOSD etiology and pathogenesis are largely unknown, a growing number of studies support the hypothesis that similar to other autoinflammatory diseases, dysregulation of inflammasome activation and the related overproduction of interleukin-1 (IL-1) β plays a pivotal role [6]. Accordingly, IL-1 blockade shows efficacy in treating AOSD symptoms in refractory cases [7]. Recent advances in sequencing technology are allowing investigators to sequence selected genes to discover low-frequency variants in patients with complex and genetically matched controls [8]. Thus, we propose that *MEFV* mutations/polymorphisms may be one of the genetic factors associated with AOSD. Therefore, in this study we investigated *MEFV* gene variations in Japanese AOSD patients.

Materials and methods

Patients

Forty eight patients diagnosed with AOSD between 2012 and 2014, and according to the diagnostic criteria of Yamaguchi *et al.* [9], were enrolled in the study from hospitals affiliated with Nagasaki University. One hundred five healthy individuals were recruited as controls. In the patient group, medical histories and clinical findings were collected by reviewing electronic medical records. Clinical, demographic, and genetic features of the 49 AOSD patients and genetic features of the 105 healthy controls were analyzed. The study protocol was approved by the ethics committees of the Sasebo City Hospital institutional review board (No 2012-A-22). Written informed consent was obtained from each individual for their clinical records to be used in this study.

Mutational analysis

Blood samples (2 ml) were collected from all subjects. Genomic DNA was extracted from whole blood using the Promega Wizard® Genomic DNA Purification Kit (Madison, WI, USA). Mutational analysis was performed by direct DNA sequencing. Polymerase chain reaction (PCR) was performed using forward and reverse primers for each exon of the *MEFV* gene, as described previously [10]. PCR products were purified using ExoSAP-IT (GE Healthcare, Tokyo, Japan) and directly sequenced using specific primers and BigDye Terminator v1·1 (Applied Biosystems, Tokyo, Japan). *MEFV* genetic analysis was approved by the Ethics Committee of Nagasaki Medical Center (No. 21003, 2009).

Immunoblot analysis

Patient's sera (1·5 μl) were diluted 10-fold with phosphate-buffered saline (PBS). We separated these diluted serum sample plus 5 μl of protein loading buffer under reducing conditions by NuPAGE 3–8% Tris-acetate gel electrophoresis (Invitrogen Carlsbad, CA, USA). Proteins were electrophoretically transferred onto an Invitrogen polyvinylidene fluoride membrane and incubated overnight at 4°C with blocking solution [5% nonfat milk in Tris-buffered saline with 0·05% Tween 20 (TTBS)]. The blocked membrane was incubated with rabbit anti-human cleaved IL-1β polyclonal antibody (MyBioSource, San Diego, CA, USA; 1:200 dilution with 1% nonfat milk in TTBS) for 1 h at room temperature and then washed five times with TTBS buffer for 10 min each time at room temperature with constant shaking. Then, the membrane was incubated with horseradish peroxidase–conjugated second antibody (1:2000 dilution; Santa Cruz Biotechnology) for 1 h at room temperature and washed five times with TTBS buffer for 10 min each time at room temperature with constant shaking. Immunodetection analysis was performed using a ECL Western blotting kit (Amersham, Little Chalfont, UK). Images of the developed film were scanned using LAS-3000 image analyzer (FUJIFILM, Tokyo, Japan).

Statistical analyses

For continuous variables, results were expressed as mean ± standard deviation (SD). For quantitative data, analysis was performed using a Mann–Whitney *U* rank-sum test to compare two independent groups. For categorical variables, a chi-square test (or Fisher's exact test when appropriate) was used for comparisons. Two-sided *P* values less than 0·05 were considered statistically significant. Data were analyzed using SPSS software (SPSS Inc., Chicago, IL, USA).

Results

Demographic features

In total, 49 AOSD patients (8 males and 41 females) were included in the study. Table 1 shows the clinical and demographic features of the AOSD patients. The mean age of patients was 51·1 ± 19·4 years (min–max: 19–84, median: 53), mean age of disease onset was 45·9 ± 20·3 years (min–max: 17–83, median: 45).

Mutational analysis

All individuals were successfully genotyped for the *MEFV* gene. *MEFV* variants were identified in 31 AOSD patients (63·3%), and the genotypes were shown in Table 2. Distributions of *MEFV* variants in patient and control groups are shown in Table 3. There was no statistical difference between AOSD patients and healthy subjects with regards allele frequencies of *MEFV* exon 1 (E84K), exon 2 (L110P, E148Q, R202Q, and G304R), and exon 3 (P369S and R408Q) variants. However, the carriage rates of exon 10 *MEFV* variants (M694I and G632S) were significantly higher in AOSD patients than those of healthy subjects (6·1% *versus* 0%, $P = 0·031$).

Clinical features in patients with or without *MEFV* variants

Clinical features of patients with or without *MEFV* variants were compared (Table 4). Although there was no statistically significant difference, AOSD patients with *MEFV* variants were more frequently treated with biologics (tocilizumab) compared with those without *MEFV* variants (19·4% *versus* 0%, $P = 0·053$). No other significant differences were found between patients with and without *MEFV* variants. However, there was a correlation between *MEFV* gene variants and AOSD disease phenotype (Table 5).

Table 1. Demographic and clinical features of adult-onset Still's disease (AOSD) patients with or without *MEFV* variants.

Demographic/clinical features	*MEFV* variants (+) ($n = 31$)	*MEFV* variants (−) ($n = 18$)	*P*-value
Age (years)	50·4 ± 18·3	52·4 ± 21·7	0·510
Onset age (years)	45·0 ± 19·8	47·4 ± 21·5	0·572
Duration (years)	5·4 ± 5·6	5·0 ± 6·2	
Gender (female/male)	25/6	16/2	0·372
Fever (%)	31 (100)	18 (100)	
Skin rash (%)	30 (96·8)	18 (100)	0·633
Sore throat (%)	21 (67·7)	14 (77·8)	0·453
Arthritis (%)	26 (83·9)	12 (66·7)	0·150
Myalgia (%)	7 (23·3)	9 (50·0)	0·058
Liver dysfunction (%)	25 (80·6)	16 (88·9)	0·372
Disseminated intravascular coagulation (%)	3 (9·7)	2 (11·1)	0·614
Macrophage activation syndrome (%)	9 (29·0)	3 (16·7)	0·270
Treatment			
Steroid treatment (%)	30 (96·8)	18 (100)	0·633
Steroid pulse therapy (%)	22 (71·0)	12 (66·7)	0·753
Immunosuppressant (%)	19 (61·3)	11 (61·1)	0·990
Biologicals (%)	6 (19·4)	0	0·053

Among clinical progression types [11], the monocyclic systemic phenotype was significantly lower in AOSD patients with *MEFV* variants. We also examined association of *MEFV* variants with AOSD treatments.

Clinical features of AOSD patients with *MEFV* exon 10 mutations

Among 49 patients with AOSD, 3 patients had exon10 mutations of *MEFV* gene. They had episodes of high fever, sore throat and arthritis. Two of them had fever-associated skin eruption colored salmon pink, and all of them were diagnosed with AOSD. In addition to oral prednisolone (12·5–40 mg/day), they were treated with methylprednisolone pulse therapy (case1), methylprednisolone pulse therapy and methotrexate (case2), and methotrexate and IL-6 inhibitor (tocilizumab) (case 3) respectively. Eventually all patients achieved remission. They were not complicated with disseminated intravascular coagulation (DIC) or macrophage activation syndrome (MAS). We previously demonstrated that the cleaved form of IL-1β, p17, was detectable in the sera from FMF patients with *MEFV* mutations [12]. We examined the circulating cleaved IL-1β in AOSD patients with *MEFV* mutations. As shown in Fig. 1, serum cleaved IL-1β bands were detectable in all AOSD patients with *MEFV* exon 10 mutations and in one of three patients with *MEFV* exon 1, 2 mutations during the febrile periods.

Table 2. *MEFV* genotypes of adult-onset Still's disease (AOSD) patients.

MEFV genotypes	AOSD $n = 49$ (%)
M694I/normal	2 (4·1)
G632S/E148Q	1 (2·0)
P369S/R408Q	3 (6·1)
E148Q/P369S	1 (2·0)
E148Q/E148Q/P369S/R408Q	1 (2·0)
L110P/E148Q/E148Q/P369S/R408Q	1 (2·0)
E148Q/normal	10 (20·4)
R202Q/normal	3 (6·1)
E148Q/E148Q	2 (4·1)
L110P/E148Q	1 (2·0)
L110P/E148Q/E148Q	2 (4·1)
L110P/E148Q/R202Q	1 (2·0)
L110P/L110P/E148Q/E148Q	1 (2·0)
E84K/normal	1 (2·0)
E84K/L110P/E148Q	1 (2·0)
Normal	18 (36·7)
Total	49

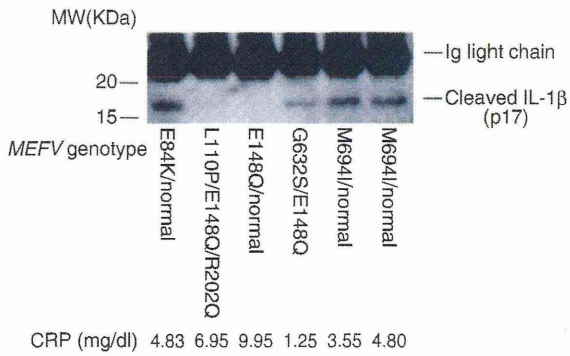

Fig. 1. Cleaved interleukin (IL)-1β (p17) in sera from adult-onset Still's disease (AOSD) patients with *MEFV* exon 10 variants or exon 1, 2 variants. Sera from AOSD patients with *MEFV* exon 10 variants ($n = 3$) or exon 1, 2 variants ($n = 3$) were analysed by anti-cleaved IL-1β immunoblot analysis. The concentrations of C-reactive protein (CRP) were shown in the lower panel of each sample. Additional brood bands observed under reducing conditions are light chains generated from cleavage of thiol-disulphide bridge of immunoglobulins.

Table 3. Allele frequencies and carriage rates of *MEFV* variants in patients with adult-onset Still's disease (AOSD).

		AOSD patients ($n = 49$)	Healthy subjects ($n = 105$)	*P*-value
Allele frequencies (%)				
Exon 10	M694I	2 (2·0)	0	0·101
	G632S	1 (1·0)	0	0·318
Exon 3	P369S	6 (6·1)	13 (6·2)	0·982
	R408Q	5 (5·1)	12 (5·7)	0·827
Exon 2	L110P	8 (8·2)	15 (7·1)	0·751
	E148Q	29 (29·6)	52 (24·8)	0·370
	R202Q	4 (4·1)	6 (2·9)	0·399
	G304R	0	6 (2·9)	0·098
Exon 1	E84K	2 (2·0)	2 (1·0)	0·380
Carriage rate (%)				
Total exon 10 variants		3 (6·1)	0	0·031
Total exon 1,2,3 variants		28 (57·1)	61 (58·1)	0·911
Total *MEFV* variants		31 (63·3)	61 (58·1)	0·542

Discussion

AOSD etiopathogenesis has not been fully elucidated but genetic and environmental factors may play critical roles [13]. Because of similarities in clinical findings between FMF and AOSD, *MEFV* gene mutations may be factors in AOSD etiology. The *MEFV* gene encodes a protein named pyrin, which is expressed in neutrophils and monocytes [14,15]. Consequently, *MEFV* gene mutations may alter pyrin function, leading to so-called 'autoinflammation' [16]. Among hereditary autoinflammatory diseases, FMF is most frequently observed. Although FMF is rare in Japan, *MEFV* mutation frequencies are relatively high [17]. In this study, we show that Japanese AOSD patients have a significantly higher frequency of disease-causing *MEFV* mutations, especially exon 10 mutations, compared with healthy subjects. FMF is characterized by recurrent fever, skin rashes, synovitis, and serositis, resembling clinical presenta-

Table 4. Demographic and clinical features of adult-onset Still's disease (AOSD) patients with or without *MEFV* variants.

Demographic/clinical features	*MEFV* variants (+) ($n = 31$)	*MEFV* variants (–) ($n = 18$)	*P*-value
Age (years)	50·4 ± 18·3	52·4 ± 21·7	0·510
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Immunosuppressant (%)	19 (61·3)	11 (61·1)	0·990
Biologicals (%)	6 (19·4)	0	0·053

Table 5. Clinical progression type of adult-onset Still's disease (AOSD) patients with or without *MEFV* variants.

Clinical progression type	Total ($n = 49$)	*MEFV* variants (+) ($n = 31$)	*MEFV* variants (–) ($n = 18$)	*P*-value	(*Pc*-value)
Monocyclic systemic (%)	15 (30·6)	5 (16·1)	10 (55·6)	0·004	(0·016)
Polycyclic systemic (%)	24 (49·0)	17 (54·8)	7 (38·9)	0·282	(n.s.)
Chronic articular monocyclic systemic (%)	6 (12·2)	5 (16·1)	1 (5·6)	0·271	(n.s.)
Chronic articular polycyclic systemic (%)	4 (8·2)	4 (12·9)	0	0·149	(n.s.)

Pc= corrected *P*-value; n.s. = not significant.

tion of AOSD. Although the function of pyrin is still unknown, it appears to inhibit processing of IL-1β to its active form [18]. With *MEFV* mutations, this action of pyrin is deficient, resulting in restricted production of active IL-1β. In this study we found that rare and low-frequency variants in an innate immunity gene, *MEFV*, were more frequently demonstrated in patients with AOSD compared to healthy subjects. The role of *MEFV* in human disease is not fully understood, however, current evidence suggests that FMF-associated *MEFV* variants are gain-of-function mutations leading to increased responsiveness to bacterial products [5]. Taken together, innate immune responses to bacterial components could be contributed to the pathogenesis of AOSD.

To date, higher frequencies of *MEFV* gene mutations have been identified in inflammatory conditions [19]. In accordance with our findings, significantly higher prevalence of the *MEFV* exon 10 variant, M694V, was found in patients with ankylosing spondylitis (AS), while no significant difference in the frequency of E148Q was demonstrated, suggesting a link between *MEFV* exon 10 rare variants and AS [20,21], a form of non-hereditary arthritis. Our data suggest that *MEFV* exon 10 variants may be one of the susceptibility factors for a complex disease, AOSD. G632S is a new mutations identified in Iranian Jews with mild phenotype of FMF [22]. G632S mutation is placed in a loop at the surface of B30·2 domain of pyrin, and this site is presumed to be functionally important [23,24].

Studies examining *MEFV* mutations in AOSD patients have previously been published in Korean and Turkish populations [25,26], both populations with high *MEFV* mutation rates. In the Turkish population, Cosan *et al.* found that although it was not significantly different, the *MEFV* gene mutation rate increased in the AOSD group [25]. However, a Korean study did not find more frequent *MEFV* mutations in AOSD patients than the general population. In this study, Kim *et al.* demonstrated higher incidence of the E148Q mutation in AOSD patients, but it was not different to healthy controls [26]. Our results on *MEFV* exon 2 and exon 3 variants are consistent with these findings. We found overall frequencies of *MEFV* variants were comparable between healthy subjects and AOSD patients. However, clinical patterns of AOSD between patients with or without *MEFV* mutations were different. *MEFV* gene polymorphisms may modify the clinical phenotype or course of inflammatory disorders [27]. We found the monocyclic systemic phenotype was significantly less common in AOSD patients with *MEFV* variants. Additionally, biologics were used more frequently in AOSD patients with *MEFV* variants. Based on these results, we suggest that being *MEFV* variants carrier may be related to repeated or sustained inflammatory processes in AOSD. Our data are inconsistent with those of Kim *et al.*, who found no association between *MEFV* exon 2 variants and AOSD clinical course [26]. These discrepancies may be owing to the gene analysis system used: we screened all exon 1, 2, 3, and 10 mutations by direct sequencing.

Our study lacked sufficient statistical power for subgroup analysis of clinical findings. Although In this study, the monocyclic systemic disease phenotype was significantly less in AOSD patients with *MEFV* variants than those without *MEFV* variants, the findings should be replicated in a large group of patients.

In conclusion, our study provides preliminary evidence suggesting that FMF-related *MEFV* variants are implicated with the disease phenotype of AOSD. Specifically, our findings suggest that *MEFV* exon 10 mutations may be one of the susceptibility factors for AOSD in the Japanese population. Replication studies are required to confirm association between these rare FMF-related *MEFV* variants with AOSD and its clinical manifestations in a larger group of patients.

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Disclosures

The authors declare that they have no competing interests.

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