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Original Investigation

The role of *MEFV* mutations in the concurrent disorders observed in patients with familial Mediterranean fever

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Abstract

Objective: This study aimed to investigate the frequency in which familial Mediterranean fever (FMF) coexists with other diseases and determine whether Mediterranean fever *(MEFV)* gene mutations are involved in such coexistence.

Material and Methods: In total, 142 consecutive patients with FMF investigated for *MEFV* mutation were enrolled in this study [Female: 87; Male: 55, mean age 32±12 years (11–62)]. All the patients were questioned for the presence of concurrent disorders, and the medical records of these patients were revised retrospectively. A previous diagnosis of inflammatory disorder other than FMF was considered true if it met the relevant criteria. *MEFV* mutations were divided into 2 groups, namely M694V and its subgroup (homozygous or heterozygous) (Group I) and others (Group II). Compound heterozygosity for M694V mutation was included in Group II to form a homogeneous group for Group I. Group I and Group II were compared according to phenotypical features. The presence of *MEFV* mutation was investigated in exons 2, 3, 5, and 10 by the multiplex-PCR reverse hybridization method.

Results: Concomitant disorders were found in 17 of 73 patients with FMF (23%) in Group I and 5 of 56 patients (8.9%) in Group II (p=0.04). Concomitant disorders in Group I were as follows: 7 cases of amyloidosis, 2 cases of Behcet's disease (BD), 4 cases of ankylosing spondylitis (AS), 1 case of antiphospholipid syndrome, 1 case of Henoch–Schonlein purpura (HSP), 1 case of combination of psoriatic arthritis, HSP, and membranoproliferative glomerulonephritis, and 1 case of AS and amyloidosis. In Group II, the following disorders were found: 1 case of amyloidosis, 1 case of BD, 1 case of AS, 1 case of ulcerative colitis, and 1 case of vitiligo.

Conclusion: The presence of M694V mutation may predispose patients with FMF to developing other inflammatory disorders.

Keywords: Familial Mediterranean fever, Behcet's disease, amyloidosis, spondylarthropathy

Introduction

Familial Mediterranean fever (FMF) is an autosomal recessive disease characterized by recurrent serositis and fever attacks. The disease is caused by mutations in a gene named Mediterranean fever (*MEFV*), which encodes a protein called pyrin. This protein is likely to have a downregulating influence on the response of neutrophils to inflammatory stimuli (1).

It has been suggested that *MEFV* mutations facilitate a heightened inflammatory response that may be beneficial against a microorganism or a specific organism (2). Nonetheless, it has been suggested that enhanced inflammation could predispose patients with FMF to some chronic inflammatory conditions. Some inflammatory disorders, including Henoch–Schonlein purpura (HSP), polyarteritis nodosa (PAN), Behcet's disease (BD), inflammatory bowel diseases, and multiple sclerosis, are also widely observed in FMF (3). *MEFV* mutation modifies not only the clinical presentation of FMF but also other inflammatory disorders. However, the role of *MEFV* mutations in FMF co-existing with other inflammatory disorders remains to be ascertained. This study aimed to investigate the frequency in which FMF coexists with other diseases and to determine whether *MEFV* mutations are involved in such coexistence.

Material and Methods

Patients

In total, 142 consecutive patients with FMF investigated for *MEFV* mutation were enrolled in this study [Female: 87; Male: 55, mean age 32±12 years (11–62)]. They were diagnosed on the basis of the Tel–Hashomer diagnostic criteria (4). The study was approved by the local ethics committee. All patients provided informed consent to participate in the study, according to the guidelines of the Declaration of Helsinki. All the patients were questioned for the presence of concurrent disorders, and the medical records of these

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Table 1. *MEFV* mutations and their distribution in 142 patients with FMF

Mutations	N	%
Group I	73	
M694V/M694V	37	23.7
M694V/-	36	23.1
Group II	56	
E148Q/E148Q	2	1.3
E148Q/-	3	1.9
V726A/V726A	1	0.6
V726A/-	3	1.9
M680I/M680I	4	2.6
M680I/-	3	1.9
M694I/E148Q	1	0.6
M680I/V726A	8	5.1
M694V/M680I	4	2.6
M680I/E148Q	4	2.6
M694V/V726A	7	4.5
M694V/R761H	6	3.8
M694I/V726A	2	1.3
M694V/K695R	1	0.6
K695R/-	1	0.6
M694V/E148Q	3	1.9
M694I/R761H	1	0.6
V726A/R761H	1	0.6
E148Q/M694V/V726A	1	0.6
Mutation (-)	13	9.1
TOTAL	142	100

MEFV: Mediterranean fever; FMF: familial Mediterranean fever

Table 2. *MEFV* mutations and concurrent disorders in FMF patients

	MEFV Mutation (+) (n=129)	Concurrent disorders
Group I (n=73)		n=17 (23%)*
	M694V	7 AMY
	-Hom	2 BD
	-Het	4 AS
		1 APS
		1 HSP
		1 PsA+HSP+MPGN
		1 AS+AMY
Group II (n=56)		n=5 (8.9%)
	Other mutations	1 AMY
		1 BD
		1 AS
		1 UC
		1 Vitiligo

*p<0.04 Group I vs. Group II

Hom: homozygous; Het: heterozygous; AMY: amyloidosis; BD: Behcet's disease; AS: ankylosing spondylitis; APS: antiphospholipid syndrome; HSP: Henoch–Schonlein purpura; PsA: psoriatic arthritis; MPGN: membranoproliferative glomerulonephritis; UC: ulcerative colitis

patients were revised retrospectively. A previous diagnosis of a disorder was considered true if it met the relevant criteria.

Mediterranean fever mutations were divided into 2 groups, namely *M694V* and its subgroup (homozygous or heterozygous) (Group I) and others (Group II). Compound heterozygosity for *M694V* mutation was included in Group II to form a homogeneous group for Group I. Group I and Group II were compared according to phenotypical features.

The severity score was calculated according to the Tel–Hashomer severity score (5). This includes mild disease (2–5 points), moderate disease (6–10 points), and severe disease (over 10 points).

The diagnosis of amyloidosis was based on histological proof of congophilic fibrillar deposits in renal biopsy specimens.

Mutation analysis

DNA was extracted from peripheral blood leukocytes using standard protocols (Invisorb® Spin Blood Kit, STRATEC Molecular GmbH, D-13125; Berlin, Germany). Molecular analyses were performed within the framework of routine genetic testing. The presence of *MEFV* mutation was investigated in exons 2, 3, 5, and 10 by the multiplex-PCR reverse hybridization method.

Statistical analysis

Statistical analysis was performed using the Statistical Package for Social Science (SPSS) software, version 15.0 statistical package program (IBM Inc.; Chicago, IL, USA). Demographic and clinical variables were summarized as proportions. Chi-square testing was performed for the comparison of categorical variables. A p value<0.05 was considered statistically significant.

Results

In total, 129 of the 142 patients with FMF had positive mutation for *MEFV* gene (90.8%), but 13 of them showed no such mutation (9.2%) (Table 1). *M694V* and its subgroup mutations (Group I) and other mutations (Group II) were found in 73 (56.6%) and 56 (43.4%) patients with FMF, respectively.

Concomitant disorders were found in 17 patients with FMF (23%) in Group I and 5 FMF (8.9%) patients in Group II (p=0.04). Concomitant disorders in Group I were as follows: 7 cases of amyloidosis, 2 cases of BD, 4 cases of ankylosing spondylitis (AS), 1 case of antiphospholipid syndrome, 1 case of HSP, 1 case of combination of psoriatic arthritis, HSP, and membranoproliferative glomerulonephritis, and 1 case of AS and amyloidosis. In Group II, the following disorders were found: 1 case of amyloidosis, 1 case of BD, 1 case of AS, 1 case of ulcerative colitis, and 1 case of vitiligo. No concurrent disorders could be found in patients with FMF having negative *MEFV* mutation (Table 2).

There was a significant difference between Groups I and II in the frequency of arthritis and erysipela-like erythema (ELE) (p<0.01 and p<0.001, respectively).

No significant difference was found between Groups I and II in the frequency of fever, abdominal pain, and pleuritic pain (p>0.05).

The number of patients with a higher severity score was significantly higher in Group I (12.9%) than in Group II (4.3%) (p<0.01). No significant differences could be found between Groups I and II in the number of patients with mild and moderate severity scores (p>0.05)

The rate of the familial history for FMF showed no significant difference in Groups I and II (63% vs. 57%, p=0.5).

Discussion

It has been reported that upregulation of the acute phase response among carriers of FMF may enhance their innate immune system and contribute to better resistance to infections (6). On the other hand, in patients with FMF, there is a subclinical inflammation even during attack-free periods, and this continued inflammation conferred by inflammatory response may predispose patients with FMF to other inflammatory disorders such as PAN and HSP (7, 8). It is well known that the presence of *M694V* allele gives rise to severe disease and amyloidosis in FMF. However, the role of *M694V* mutations in the development of concurrent disorders in the course of FMF is not well known. Kalyoncu et al. (9) showed that patients who are carriers for *MEFV* mutations may have a tendency to develop musculoskeletal rheumatologic complaints. Moreover the presence of these mutations may affect their disease course when they develop other rheumatic diseases.

We included Group I patients having only *M694V* allele (homozygote or heterozygote) to form a homogenous group. The patients with compound heterozygosity for *M694V* were added to Group II. We considered that this stratification may be a better way to determine whether *M694V* mutations play a role in concurrent disorders seen in patients with FMF. In line with the literature, the patients with *M694V* mutations had a severe disease score. Arthritis and ELE were more commonly determined in Group I. Seventeen patients having M694V mutations had concurrent disorders.

The most common concurrent disorder apart from amyloidosis in patients with FMF was AS. Six of 142 patients with FMF (4.2%) had AS. The frequency of AS in Group I was 6.8%. In Group II, AS was found at a frequency of 1.7%. We have previously that 7.3 of 256 patients with FMF had sacroiliitis, which was related to the presence of *M694V* mutations (10). Akar et al. (11) showed that patients with FMF having sacroiliitis more commonly had *M694V* mutation compared than those without sacroiliitis.

Kaşifoğlu et al. (12) reported that 8.6% of 2296 Turkish patients with FMF had amyloidosis and amyloidosis was associated with male gender, arthritis, and the presence of *M694V* genotype. We detected amyloidosis in 9 of 142 (6.3%) patients with FMF.

While amyloidosis was determined only 1 case with FMF in Group II (1.78%), the frequency of amyloidosis in Group I was found to be 10.9%.

The relationship between BD and FMF has been debatable for a long time. Chetrit et al. (13) found 2 patients with BD among 355 patients with FMF and 2 patients with FMF among 53 patients with BD. Sixteen patients with BD were found to have *MEFV* mutations, and 2 of them had FMF. Based on these results, they suggested that the association between BD-FMF was higher than expected in both directions. However, the small number of patients was a matter of concern in this study. Bakkaloğlu et al. (14) evaluated the presence of associate diseases in 2838 Turkish patients with FMF, and BD was found in 0.5% of patients with FMF. In our study, in Group I, we found 2 patients with FMF accompanied by BD, which is little high compared with the number reported in the literature. In Group II, only one patient with FMF had BD.

It is well known that the presence of *MEFV* mutation in cases without FMF can modify clinical manifestations of that disease. Atagündüz et al. (15) showed that there is a tendency to develop vascular involvement in BD patients with *MEFV* mutation.

The association of PAN and HSP with FMF is well known. The prevalence of HSP and PAN in Turkish patients with FMF has been found to be 7% and 1%, respectively (15). The prevalence of HSP in the general population has been found to vary from 0.05% to 0.8% (8, 16). In that study, genotype analysis was not conducted. Tekin et al. (16) investigated the role of *MEFV* mutations in 11 patients with HSP. They could not find specific *MEFV* mutations associated with FMF-HSP. In our study, we did not find any patient with PAN. HSP was found in 2 of 142 (1.4%) patients with FMF. These 2 patients had *M694V* mutations. It is difficult to draw a concrete conclusion from these results because of the small number of patients.

Inflammatory bowel diseases are possibly associated with FMF. Fidder et al. (17) determined 7 patients with Crohn's disease among 4978 FMF patients. Of 6 patients determined as having *MEFV* mutations, 3 had homozygosity for *M694V*, 1 had compound heterozygosity for *M694V/V726A*, and 1 had heterozygosity for *M694V*. One patient had no mutation. Yildirim et al. (18) investigated the prevalence of *MEFV* mutations in patients ulcerative colitis. The mutations were identified in 19 of the 54 (35.2%) patients with ulcerative colitis. Homozygous *E148Q* was determined in 2 patients (3.7%) and heterozygous *E148Q* in 17 patients (31.5%) (*E148Q* 11.1%, *M694V* 5.6%, *V726A* 5.6%, *K695R* 1.8%, *M680I* 1.8%, and compound heterozygous 5.6%). In our study, we found 1 UC patient in Group II.

Bias is inevitable in our study because the presence of concomitant disorders significantly increases the chance of a disease being detected (Berkon's bias). The inclusion of disease controls in concurrence studies minimizes the impact of this important bias at least to some degree (19). We could not include a disease control group in this study because of financial problems. The other limitation is that we formed Group 1 from only patients with FMF having homozygosity and heterozygosity for *M694V*. Compound heterozygosity for *M694V* was included in Group II. This categorization was made to form a homogenous group.

In conclusion, compared with FMF patients with other mutations, concurrent disorders were frequently determined in patients with FMF having *M694V* mutations and its subgroups. The patients with *M694V* mutation and its subgroups (homozygous, heterozygous) more commonly had arthritis, ELE, and a higher severe severity score than those with other mutations. *MEFV* mutations, particularly *M694V* mutations, can modify both clinical manifestations of FMF itself and those of other inflammatory disorders. The presence of *M694V* mutation in addition to other genetic and environmental factors may predispose FMF patients to developing other inflammatory disorders.

Ethics Committee Approval: Ethics committee approval was received for this study from the ethics committee of Eskişehir Osmangazi University School of Medicine (21 May 2010/119).

Informed Consent: Written informed consent was obtained from patients who participated in this study.

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