MEFV mutations are increased in Behçet's disease (BD) and are associated with vascular involvement

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ABSTRACT

Objective. *A high prevalence of Behçet's disease (BD) among familial Mediterranean fever (FMF) patients has been described recently and a weak association of BD and certain MEFV gene mutations, originally linked to FMF, has been reported in an ethnically mixed population from France. We further investigated the presence of MEFV mutations in BD patients from Turkey, a country with a high prevalence of both disorders.*

Methods. *The frequencies of three FMF-related MEFV mutations (M694V, M680I and V726A) were investigated in BD patients (n = 57) by molecular genetic studies using a polymerase chain reaction with the ARMS method. All patients fulfilled the International Study Group Criteria for the diagnosis of BD and patients with FMF-like symptoms or a chronic inflammatory disease were excluded.*

Results. *Fifteen BD patients were found to carry one single MEFV mutation (26%), compared to 9.1% in the control group (p = 0.003, OR:3.5, 95% CI: 1.6-7.6). Among 20 BD patients with vascular involvement, 11 (55%) had MEFV mutations compared to 4 patients (11%) in the non-vascular group (p = 0.001, OR: 10, 95% CI: 2.5-39.3). M694V was the dominant mutation in our study group (11 out of 15 patients with mutated alleles). Six out of 7 female patients with vascular involvement carried MEFV mutations in contrast to 5 out of 13 male patients (85.7% versus 38.4%, p = 0.07, OR: 0.1, 95%CI: 0.009-1.14). No association with other clinical manifestations was observed.*

Conclusion. *MEFV mutations, originally linked to FMF, may act as a genetic susceptibility factor for other inflammatory disorders such as vascular BD.*

Introduction

Mutations of the MEFV gene, which encodes the neutrophil protein pyrin (marenostrin) was recently linked to familial Mediterranean fever (FMF), an autosomal recessive, inflammatory disease characterized by recurrent episodes of febrile serositis attacks. Behçet's disease (BD) is a multi-systemic vasculitic disorder with mucocutaneous,ocular, arthritic, vascular and central nervous system involvement. A clinical relationship between FMF and BD has been described in Israel, with a slightly higher prevalence of BD among FMF patients compared to the general population (1). Recently, a weak association of certain MEFV mutations and BD has also been reported in an ethnically mixed population from France (2). We further investigated the presence of MEFV mutations in BD patients from Turkey, a country with a high prevalence of both BD and FMF.

Materials and methods

Patients

We investigated the presence of MEFV mutations in BD patients from Turkey (n = 57, 26 males, 31 females, mean age 38 ± 11 years) recruited from the Behçet's disease outpatient clinics of Marmara Medical Faculty, Istanbul. All BD patients had oral ulcers. Genital ulcers were present in 86%, skin manifestations in 75%, ocular involvement in 21% (all posterior uveitis except for one patient) and vascular involvement in 35%. The pathergy test was positive in 53% of the study patients. At the time of the study, 23% of the patients were receiving immunosuppressive treatment. The study was approved by the University of Marmara Institutional review board and informed consent was obtained.

Inclusion and exclusion criteria

Consecutive patients were screened for

Table I. The distribution of MEFV mutations in BD patients

Patient no.	Mutation	Oral ulcer	Genital ulcer	Positive pathergy test	Uveitis	Cutaneous signs	Venous involvement	Treatment
BD patients with vascular involvement								
3	M694V / M694V	YES	YES	YES	NO	EN	DVT, V. Cava inf. syndrome	AZA, MP, Warfarin
6	M680I / -	YES	YES	YES	NO	NO	DVT	COL, ASA
13	M694V / -	YES	YES	YES	NO	EN	BC syndrome	AZA, MP, Warfarin
14	V726A / -	YES	YES	YES	YES	EN	DVT, BC syndrome	AZA, MP, Warfarin
20	M694V / -	YES	NO	YES	YES	NO	DVT, BC syndrome	CYC, AZA, MP
24	M694V / -	YES	NO	YES	NO	EN	DVT, CNS involvement	AZA, Warfarin, COL
33	M694V / -	YES	YES	YES	YES	EN	Nodular vasculitis	COL
34	M694V / -	YES	YES	NO	NO	EN	DVT, ST	COL
53	M694V / -	YES	YES	NO	NO	EN	DVT	COL
54	M694V / -	YES	YES	NO	NO	EN	DVT	COL, NSAID
57	M694V / -	YES	YES	YES	NO	EN	ST	COL
BD patients without vascular involvement								
25	V726A / -	YES	YES	YES	NO	EN	NO	COL, NSAID
29	M694V / -	YES	YES	NO	NO	EN	NO	COL, NSAID
30	V726A / -	YES	YES	YES	YES	EN	NO	COL, NSAID
38	M694V / -	YES	YES	YES	NO	EN	NO	COL, NSAID

ASA: Acetyl salicylic acid; AZA:Azathiopirin; BC Synd: Budd-Chiari syndrome; CNS: Central nervous system; COL: Colchicum dispert; CYC: Cyclophosphamide; DVT: Deep vein thrombosis; EN:Erythema nodosum; MP: Methylprednisolone; NSAID:Non-steroid anti-inflammatory drugs; ST: Superficial thrombophlebitis

the diagnostic criteria of the International Study group for BD and the Tel-Hashomer criteria for FMF (3, 4). Patients fulfilling the Tel-Hashomer criteria for FMF or patients with symptoms resembling those of FMF were excluded. Patients with a chronic inflammatory disease other than FMF also were excluded from the study. A non-inflammatory group of both healthy (n = 50, M/F 23/27) and disease controls (n = 136, M/F 62/74) with cardiovascular system disorders (n = 48), metabolic disorders (n = 58), renal disease (n = 16), or pulmonary disease (n = 14) were also studied. Mutational analysis with PCR was done according to a protocol described earlier by Eisenberg *et al*. (5).

Statistical analysis

The allele frequencies were compared by the Fisher's exact test.

Results

Fifteen BD patients were found to carry one single MEFV mutation (26%), compared to 9.1% in the control group, with a statistically significant difference between the groups (p = 0.003, OR:3.5, 95% CI:1.6-7.6). All patients were heterozygous for MEFV mutations except for one who was homozygous for M694V (Table I). M694V was the dominant mutation in our study group. Among 20 BD patients with vascular involvement, 11 (55%) had MEFV mutations compared to 4 patients (11%) in the non-vascular group (p = 0.001, OR:10.0, 95% CI:2.5-39.4). The allele frequencies in the non-vascular group did not differ from the control group (11% vs. 4.5%) (p = 0.76).

All four patients with severe vascular involvement (Budd-Chiari syndrome, vena-cava superior thrombosis and vascular neuro-BD) were mutation positive (p = 0.004, OR: 33.2, 95% CI: 1.6-664.2). MEFV mutations were also more frequent in the pathergy positive group (40% vs. 11%,p = 0.02, OR: 5.3, 95% CI: 1.3-21.7). The frequency of mutations did not differ in patients with uveitis (31% vs. 25%, p = 1.0, OR: 1.2, 95%, CI: 0.3 – 4.7) and no association between other clinical manifestations of BD and MEFV mutations was observed. Six out of 7 female patients with vascular involvement carried at least one MEFV mutation compared to 5 out of the 13 male patients (85.7% vs. 38.4%, p = 0.07, OR:0.1, 95% CI: 0.009 – 1.14). Similar to two recent reports by Yilmaz *et al.* and Ozdogan *et al*. we found a carrier rate of 9% for the M694V, M680I and V726A mutations in the Turkish population (6, 7).

Discussion

Epidemiological similarities and the neutrophilic nature of the inflammatory infiltrates suggest that a common factor may be present in BD and FMF (8). Touitou *et al.* first suggested the possible implication of MEFV mutations in BD, reporting slightly higher frequencies of M694V, V726A and E148Q mutations (2.6%, 2.6%, and 5.2%, respectively) compared to the controls (0%, 0%, and 2.2%) (2). However, Touitou *et al*. studied BD patients with diverse ethnical backgrounds, possibly including some from populations with low MEFV penetration. Only 6 patients were of Turkish origin in their study and 2 of them were positive for MEFV mutations. On the other hand, a similar frequency of MEFV mutations to our study among BD patients (30%) has been reported by Ben-Chetrit *et al*. from Israel (9). An association of MEFV mutations with the severity of rheumatoid arthritis and multiple scle-

rosis has also been reported recently, suggesting that MEFV mutations might be a severity marker for other inflammatory disorders, a question not addressed by the previous studies (9-12).

Although vascular complications are an exception in the course of FMF, an important observation of our study is the high prevalence of MEFV mutations in BD patients with vascular involvement. Severe vascular complications such as Budd-Chiari syndrome, vena-cava superior thrombosis and vascular neuro-BD were found to be present only in the mutation positive group of BD patients. Factor-V Leiden and prothrombin 20210 mutations have also been linked to vascular thrombosis in BD previously (13). However, whether the increased propensity to thrombosis in BD is intra-vascular or linked to the endothelial damage is still controversial. Recently, neutrophilic infiltration of the *vasa vasorum* has been described as the main pathologic feature in the vascular specimens of BD patients (14). Although controlled studies are not available, immunosuppressives are also the first choice treatment for vascular BD by most experts (15).

Male BD patients are described to have a more severe clinical course and vascular involvement is also more common in male patients (13/26 versus 7/31) in our study (16). However, the higher prevalence of MEFV mutations among female patients with vascular complications (6/7 versus 5/13) suggests that the presence of the MEFV gene might be a more crucial genetic factor in female patients for vascular involvement. Ben-Chetrit *et al*. did not report any association of MEFV mutations with the clinical manifestations of BD or gender (9). However, there was only one patient of Turkish origin in their study and the differences between the two studies could be explained by the differences in ethnical background.

In conclusion MEFV mutations, especially of M694V, may act as another genetic susceptibility factor, especially for vascular BD, in Turkish BD patients. Further studies evaluating the role of MEFV mutations in BD patients from other ethnic populations and on different neutrophilic disorders such systemic neutrophilic vasculitides will further help to elucidate the exact role of MEFV gene on neutrophil-related inflammatory mechanisms.

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References

1. SCHWARTZ T, LANGEVITZ P, ZEMER D, GAZIT E, PRAS M, LIVNEH A: Behçet's disease in Familial Mediterranean fever:Characterization of the association between the two diseases. *Semin Arthritis Rheum* 2000; 29: 286-95.
2. TOUITOU I, MAGNE X, MOLINARI N *et al*.: MEFV mutations in Behçet's disease. *Hum Mutat* 2000; 16: 271-2.
3. INTERNATIONAL STUDY GROUP FOR BEHÇET'S DISEASE: Criteria for diagnosis of Behçet's disease. *Lancet* 1990;335:1078-80.
4. LIVNEH A, LANGEVITZ P, ZEMER D *et al*.: Criteria for the diagnosis of familial Mediterranean fever. *Arthritis Rheum* 1997; 40: 1879-85.
5. EISENBERG S, AKSENTIJEVICH I, DENG Z, KASTNER DL,MATZNER Y: Diagnosis of Familial Mediterranean Fever by a molecular genetics method. *Ann Intern Med* 1998; 129: 539-42.
6. YILMAZ E, OZEN S, BALCI B *et al*.: Mutation frequency of familial Mediterranean fever and evidence for a high carrier rate in the Turkish population. *Eur J Hum Genet* 2001; 9: 553-5.
7. OZDOGAN H, SAYMAN N, MELIKOGLU M *et al*.: MEFV gene mutations in Turkish patients with FMF amyloidosis versus other secondary amyloidosis. *Clin Exp Rheumatol* 2000; 18: Abst. E-5: 297.
8. DIRESKENELI H: Review: Behçet's disease: Infectious etiology, new autoantigens and HLA-B51. *Ann Rheum Dis* 2001; 82: 996-1002.
9. BEN-CHETRIT E, COHEN R, DHAJEK-DHAUL T: Familial Mediterranean fever and Behçet's disease – Are they associated ? *J Rheumatol* 2002; 29: 530-4.
10. RABINOVICH E, LIVNEH A, LANGEVITZ P, SHINAR E,ZAKS N, SHINAR Y: Mediterranean Fever gene (MEFV) mutations and rheumatoid arthritis:A severe combination. *Clin Exp Rheumatol* 2002; 20 (Suppl. 26): A16, S-73 (abstract).
11. SHINAR Y, LIVNEH A, VILLA Y, PINHASOV A, ZEITUN I, ACHIRON A: MEFV mutations and autoimmune diseases:A severe combination. *Clin Exp Rheumatol* 2002; 20 (Suppl. 26): A28, S-75 (abstract).
12. BEN-CHETRIT E, YAZICI H: Thoughts on the proposed links between Behçet's disease and familial Mediterranean fever. *Clin Exp Rheumatol* 2002; 20 (Suppl. 26): S1-S2.
13. GUL A, ASLANTAS AB, TEKINAY T, KONIÇE M, OZCELIK T: Procoagulant mutations and venous thrombosis in Behçet's disease. *Rheumatology* 1999; 38: 1298-99.
14. KOBAYASHI M, ITO M, NAKAGAWA A *et al*.: Neutrophil and endothelial cell activation in the *vasa vasorum* in Behçet's disease. *Histopathology* 2000; 36: 362-71.
15. BARNES CG, YAZICI H: Editorial. Behçet's syndrome. *Rheumatology* 1999; 38: 1171-76.
16. YAZICI H,TÜZÜN Y, PAZARLI H *et al*.: Influence of age of onset and patient's sex on the prevalence and severity of manifestations of Behçet's syndrome. *Ann Rheum Dis* 1984; 43: 783-9.