Türkiye Parazitoloji Dergisi, 33 (2): 158 - 161, 2009
© Türkiye Parazitoloji Derneği

Türkiye Parazitol Derg.
© Turkish Society for Parasitology

What Is The Importance of *Demodex folliculorum* in Behçet's disease?

Sinan EMRE[1], Ozlem Makbule AYCAN[2], Metin ATAMBAY[2], Semsettin BILAK[3], Nilgun DALDAL[2], Yelda KARINCAOGLU[4]

[1]Celal Bayar University Medical Faculty Department of Eye Diseases, Manisa, [2]Inonu University Medical Faculty Department of Parasitology, Malatya, [3]Inonu University Medical Faculty Department of Eye Diseases, Malatya, [4]Inonu University Medical Faculty Department of Dermatology, Malatya, Turkey

SUMMARY: *Demodex folliculorum* is an obligate parasite and commonly detected in patients with immune system deprivation. This study is planned to document the *Demodex folliculorum* prevalence among patients with Behçet's disease (BD). The patients who referred to the ophthalmology clinic were included in the study. Fourty patients with BD and 131 patients with refractive errors without any ocular and systemic disease were included. For parasite detection, 3 eyelashes from each inferior eyelid were epilated. Standardized skin surface biopsy (SSSB) was performed for detection of parasite at cheeks of patients. Samples were prepared with Hoyer's solution and investigated under the light microscope. There were 15 female and 25 male in BD group and 61 female and 70 male patients ine control group. Mean ages were 37.62 and 38.38 for BD and control groups, respectively. *Demodex folliculorum* prevalence at eyelashes was 65% for BD and 10% for control group. SSSB of cheek revealed 7.5% positivity for BD and 10% for control group patients. Statistical analysis documented a significant difference for eyelashes ($p<0.05$) which could not be detected for skin results. Investigation of *Demodex folliculorum* in BD may be useful, even in patients without any complaint, for the treatment of ocular and eyelid dyscomforts of these patients.

Key Words: Behçet's disease; *Demodex folliculorum*; eyelash; prevalance

Demodex folliculorum'un Behçet Hastalığındaki Önemi Nedir?

ÖZET: *Demodex folliculorum*, insan pilosebase bezlerinin zorunlu bir parazitidir ve sıklıkla immun sistem yetmezliği bulunan kişilerde tespit edilmektedir. Bu çalışmanın amacı, *Demodex folliculorum* sıklığını Behçet hastalığı bulunan hastalarda araştırmaktır. Çalışmaya İnönü Üniversitesi Tıp Fakültesi Göz hastalıkları Anabilim dalında takipleri yapılan ve Dermatoloji Anabilim dalınca refere edilen 40 Behçet hastası dahil edilmişlerdir. Refraksiyon problemleri dışında sistemik ve oküler herhangi bir hastalığı olmayan 131 hasta ile kontrol grubu oluşturulmuştur. Parazit tespiti için, her bir alt kapaktan üçer adet kirpik epile edilmiştir. Ayrıca hastaların yanak yüzlerinde parazit tespiti için standart cilt yüzey biyopsisi (SCYB) uygulanmıştır. Epile edilen kirpikler ve cilt testleri Hoyer solusyonu uygulanarak, x100 büyütmeli ışık mikroskobunda incelenmişlerdir. Çalışma grubu 15 bayan, 25 erkek Behçet hastasından, kontrol grubu 61 bayan, 70 erkek hastadan oluşmaktaydı. Grupların ortalama yaşları sırasıyla 37.62 ve 38.38 idi. Behçet hastalarında kirpik diplerinde *Demodex folliculorum* sıklığı % 65, kontrol grubunda % 10'du. Cilt SCYB sonuçlarına göre *Demodex folliculorum* sıklığı Behçet hastalarında % 7.5, kontrol grubunda % 10'du. İstatistiksel analiz, kirpik dipleri için farkın anlamlı olduğunu ($p<0.05$) ancak yanak yüzeyleri için anlamlı olmadığını ortaya koymuştur. Behçet hastalarında *Demodex folliculorum* sıklığının araştırılması, şikayetleri olmasa bile bu hastalarda oküler yüzey ve gözkapağı problemlerinin tedavisinde faydalı olabilir.

Anahtar Sözcükler: Behçet hastalığı; *Demodex folliculorum*; kirpik; prevelans

INTRODUCTION

The hair follicle mites *Demodex folliculorum* and *Demodex brevis* are the most common permanent ectoparasites of man which asymptomatically parasitizes normal skin. These mites are spindle-shaped, their body surface is hairless and colourless. They have 0.3-0.4 mm length and four short pairs of legs. The greatest concentration is found in body sites where sebaceous glands are numerous and sebum production is pronounced. Main habitats are the hair follicles and sebacesous glands of the facial skin (31). Over the facial skin parasites have been determined on the cheeks and forehead including nose, temples, chin and neck (18). *Demodex folliculorum* mites are found in the infundibular portion of hair

Makale türü/Article type: **Araştırma / Original Research**
Geliş tarihi/Submission date: 19 Kasım/19 November 2008
Düzeltme tarihi/Revision date: 06 Şubat/06 February 2009
Kabul tarihi/Accepted date: 06 Şubat/06 February 2009
Yazışma /Correspoding Author: Sinan Emre
Tel: (90) (312) 202 46 25 Fax: (90) (312) 214 11 31
E-mail: mdsinanemre@yahoo.com
It was presented in the 9th International Ocular Inflammation Society Congress (September 17 -20, 2007, Paris-France)

follicles while *Demodex brevis* burrow deeper into the sebaceous glands and ducts (5).

Demodex infestation in humans have been described in several clinical forms as; rosacea-like demodicosis, pityriasis folliculorum, perioral dermatitis and blepharitis (4, 18, 31). *Demodex* blepharitis has been first described by Raehlmann in 1898. After that several researchers reported presence of parasites in lash follicles at various frequencies in different study groups (8, 13, 20).

The reported rate of parasite carriers among healthy subjects varies and may increase upto 100% symptoms are mainly developed in people with predisposing factors (29). Association of increased frequency of demodicosis with immune system dysfunctions support this idea (7, 15-17, 19, 23, 28). When the mite multiplies and reaches to a sufficient number, it can become pathogenic due to its enhanced irritating action (12). The host immune defence appears to be the most important factor to prevent mite overgrowth.

Behçet's disease is a chronic, relapsing, multisystemic idiopathic inflammatory disease, with classical symptoms consisting of oral aphthae, genital ulcers, and uveitis. This ubiquitous disorder exhibits a distinct geographic variation and is endemically higher especially in Turkey, Iraq, Iran, Korea and Japan, countries placed on the ancient Silk Road. It accounts for up to 20% of cases of endogenous uveitis in some of these countries, such as in Japan and Turkey and the highest prevalence is reported in Turkey where family occurrence has been noted (10). In the etiopathogenesis of BD so many different factors have investigated up to now. But intermittent nature of the disease and the lack of consistent response to therapy make the underlying etiology difficult to define. Probably it is mediated by combination of genetics, infectious agents, immune dysregulation and inflammatory mediators, shock proteins, oxidative stress, lipid peroxidation, and environmental factors (10). Studies have shown that both cellular and humoral immune system has been involved in etiopathology of Behçet's disease (10, 24).

To our knowledge, the prevalence of *Demodex folliculorum* in Behçet's disease patients have not been investigated up to date. The purpose of this study was to investigate the prevalence of *Demodex folliculorum* at eyelashes and cheek skin among patients with Behçet's disease and control group composed of patients without any systemic and ocular disease except simple refractive errors.

MATERIALS AND METHODS

The patients diagnosed as Behçet's disease at dermatology clinic were referred for ocular findings and the patients who were first applied for ocular complain who were diagnosed as Behçet's disease after appropriate consultations, were included. International Behçet's Study Groups criteria were used for diagnosis of Behçet disease. There were 40 patients with Behçet's disease (25 male and 15 female). Among 40 Behçet's disease patients 19 were newly diagnosed patients and were free of any topical and systemic sterois and/or immunosuppresive treatment. On the other hand, 21 patients were under systemic immuno-modulatuar treatment with at least one form of medication. The patients with blepharitis and patients under any type of treatments were excluded for avoidance the effect of comorbidity and medications. Control group was composed of 131 (70 male and 61 female) patients with just refractive errors without any other ophthalmologic and systemic disease.

For *Demodex* detection, under slit-lamp biomicroscope 3 eyelashes from each inferior eyelid were epilated with a fine forceps. Eyelashes were placed on glass slide and were mounted with a coverslip. Standardized skin surface biopsy (SSSB) was preffered for detection of parasite at cheeks of patients. Epilated eyelashes and cheek samples were prepared with Hoyer's solution for investigation under light microscope with x100 magnification. For eyelashes determination of 3 or more living parasites at the root of each eyelash was diagnosed as infestation. For skin samples infestation, five of more living parasites in a 1 cm^2 was requiered.

To be included in the study, informed consent was obtained from all participants. The study was approved by the ethical committee of Inonu University School of Medicine and carried out in accordance with the Declaration of Helsinki.

Statistical analysis were performed with SPSS for Windows version 12.0 program (SPPS Inc., Chicago, IL). All data were reported as means ± standard deviation (SD). Pearson Chi square test was used for statistical analysis of eyelash results. For comparison of skin results Fisher exact chi square test was used. A value of $p< 0.05$ was considered statistically significant.

RESULTS

The 40 Behçet's disease cases comprised 25 males (62.5%) and 15 female (37.5%). Mean age of Behçet's disease group was 37.62 ± 9.47 years (range 21- 57 years). Meanwhile mean age of control group was 38.29 ± 17.69 years (range 12-84). *Demodex folliculorum* was detected at eyelashes in 26 of 40 (65%) Behçet's disease patients, but in only 13 of 131 (10 %) healthy controls. The difference in mite prevalence was statistically significant ($p <0.0001$). The rate of *Demodex folliculorum* detection at cheek of Behçet's disease patients was 7.5% (3/40), but at control group it was 10% (13/131). Statistical analysis revealed an unsignificant difference between groups ($p>0.05$). The demodicosis incidence according to study groups were shown in Table 1.

Table 1. *Demodex folliculorum* incidence among study groups.

	Behçet's Disease	**Control group**
***Demodex folliculorum* + at eyelashes (%)**	26/40 (65 %)	13/131 (10 %)
***Demodex folliculorum* + at cheeks (%)**	3/40 (7.5 %)	13/131 (10 %)

DISCUSSION

The greatest concentration of *Demodex folliculorum* is found in body sites where sebaceous glands are numerous and sebum production is pronuonced. The most common site of the parasite is the face and more specifically the cheeks, and forehead including nose, nasolabial folds, temples and chin (5, 18, 31). *Demodex folliculorum* consumes epithelial cells, produces follicular distention, hyperplasia and leads cuffing in eyelashes secondary to increased keratinization. Demodectic mites histologically cause inflammatory changes, epithelial hyperplasia and follicular plugging. At histologic sections of lid follicles infested with *Demodex folliculorum*, distention and thickening were observed. Easier epilation of eyelashes is the result of follicular inflammation. Also eyelashes becomes more brittle in the case of demodicosis. Clinically collar around the base of the lashes and madarosis are possible findings (www.emedicine.com/oph/topic517htm). According to Gao et al, lashes with cylindrical dandruff are pathognomonic for ocular *Demodex* infestation (13). On the other hand, *Demodex* species-induced pathologic changes have been reported in dry eye conditions (14). Infestation of meibomian or Zeis gland causes reduction of the superficial lipid layer of the tear film. Also parasitic involvement of meibomian gland has been reported to cause chalazion (26). Recently, Kheirkhah et al decribed the corneal manifestations secondary to ocular *Demodex* infestation. The authors clearly documented the regression of corneal superficial vascularization, improvement of phlyctenule-like lesion and marginal corneal infiltrations after treatment of demodicosis (21).

The high rate of parasite in our study group may also be related to the medications of patients. In our study group 19 patients were free of medications (47.5%). Most of these patients were have newly diagnosed BD under investigation. From these patients 14 had (73.6%) positive results for *Demodex folliculorum* at their eyelashes. On the other hand, 21 of our patients were need medication for control of Behçet's disease. Among this group the rate of *Demodex folliculorum* at eyelashes was 57% and was lower than the patients without any medication. At that point it was interesting to get lower results from patients who were taking various types immunomodulators. It seems that immune status of the patient may not be only factor for mite infestation. In our study group patients under medical control were taking several combinations of medications, for that reason the number of patients at each medication combination were very limited for making any statistical analysis.

Primary or secondary immunodepression may be the factors for transition from a clinically unapparent colonization of mites to dermatosis (2). Primary immunodepression is most probably based on a hereditary defect of T cells, subsequently reinforced by substances that are produced by mites. It was shown that mites produce a humoral factor, which causes selective supression of T lymphocytes and this factor blocks the local immune response (2). Several studies showed that most T cells in the dermal granulomatous infiltrates around *Demodex* parts, were helper/inducer T cells (14, 30). The predominance of CD4 helper T cells in the dermal infiltrates of lesions associated with demodicosis, suggesting that a cell-mediated immune response has an important role in the pathogenesis of demodicosis (2). Meanwhile Georgala et al put forward a new hypothesis that a delayed hypersensitivity reaction (type IV immune response) to an unknown antigen, could occur in *Demodex* infestation (14). Akilov and Mumcuoglu hypothezied that NK2 cells are the responsible for the elimination of *Demodex* mites and that the disintegrating parts of the mites cause the activation of this lymphocyte subpopulation. Also in the presence of Cw2 or Cw 4 phenotypes, the killing activity is directed to body's own T lymphocytes (2). However, the immunocompromised condition of the patient might not only be a predisposing factor for mite infestation, but could also deteriorate during mite parasitism. It was shown that T cells are the major target of mite immuno-suppression. During mite infestation the quantity of T lympho-cytes was reduced, but mature B cells remained unaffected (2).

Secondary immunosuppression may be stimulating mechanism in clinical manifestation of demodicosis particularly following corticosteroid or chemotherapy or due to diseases of an immunocompromised nature such as leukemia and AIDS (9-14). Recently a study documented generalized demodicosis in 19% of 56 children with acute lymphoblastic leukaemia who were receiving chemotherapy (16). Also there are reports of refractory demodex folliculitis in children with acute lymphoblastic leukemia (15). The association of demodicidosis with acquired immunodeficiency syndrome has been reported several times (7, 17). But in contrast to this, there are also reports of demodicidosis in healthy young children (25). On the other hand, *Demodex folliculorum* could not be find any of 30 renal transplant patients receiving combination therapy of cyclosporin, azathioprine and prednisolone (3). Meanwhile, a study planned in our institute documented statistically significant difference between end-stage chronic renal patients and controls (19). This rise the question of what are the factors other than generalized immune suppression leading to the development of demodicosis.

Topical immune supression with steroid may be an other cause of demodicosis. It has been shown that topical steroids can cause rosacea-like lesion on previously healthy skin (9) or may exagerate symptoms of demodicosis (27). Different studies reported increased *Demodex* population in rosacea group compared to control group (5, 6, 22). But most marked increase was described in those with steroid-induced rosacea (5, 6).

The *Demodex* prevelance in chronic blepharitis have been extensively studied. Demmler et al., reported the prevalence of *Demodex* 52% in patients with chronic blepharitis and also authors reported high gram positive and negative bacteria association in patients with *Demodex* (8). In contrast to this report, Arıcı et al. did not find any difference for the prevalence of *Demodex folliculorum* in blepharitis patients and control group. Also they did not find any change with host factors such as age

and gender (20). Also Akilov et al found that 21.8% of patients eyelids and eyelashes were affected by mites in combination with skin lesions due to demodicosis (1). The proliferation of *Demodex* inside the meibomian glands could be responsible for the meibomian gland dysfunction observed in rosacea and therefore of the secondary ocular surface impairment (11, 21). In our study group, the prevalence of *Demodex folliculorum* at eyelashes of control group was 10% and this rate was lower then the reported rate from one of our neighbour city (20).

Demodex folliculorum is an inhabitant of pilosebaceous follicles and has been implicated in rosacea, blepharitis and a variety of ocular surface pathologies. The high prevalence rate of this study showed that during the treatment of intraocular inflamation ophthalmologists also should pay attention to ocular surface inflamations of these patients and *Demodex folliculorum* should be taken care in patients with any sign and symptom related to ocular surface pathologies.

REFERENCES

1. **Akilov OE, Butov YS, Mumcuoglu KY,** 2005. A clinico-pathological approach to the classification of human demodicosis. *J Dtsch Dermatol Ges*, 3: 607-614.
2. **Akilov OE, Mumcuoglu KY**, 2004. Immune response in demodicosis. *J Eur Acad Dermatol Venereol*, 18: 440-444.
3. **Aydıngöz IE, Mansur T, Dervent B**, 1997. *Demodex folliculorum* in renal transplant patients. *Dermatology*, 195: 232-234.
4. **Baima B, Sticherling M**, 2002. Demodicosis revisited. *Acta Derm Venerol*, 82: 3-6.
5. **Basta-Jusbasic A, Subic JS, Ljubojevic S**, 2002. *Demodex folliculorum* in development of dermatitis rosaceiformis streroidica and rosacea-related diseases. *Clin Dermatol*, 20: 135-140.
6. **Bonnar E, Eustace P, Powell FC**, 1993. The *Demodex* mite population in rosacea. *J Am Acad Dermatol*, 28: 443-448.
7. **Delfos NM, Collen AF, Kroon FP**, 2004. Demodex folliculitis: a skin manifestation of immune reconstitution disease. *AIDS*, 18: 701-702.
8. **Demmler M, Kaspar HM, Möhring C, Klauss V,** 1997. Blepharitis. *Demodex folliculorum*, associate pathogen spectrum and specific therapy. *Ophthalmologe*, 94: 191-196.
9. **Egan CA, Rallis TM, Meadows KP, Krueger GG**, 1999. Rosacea induced by beclomethasone dipropionate nasal spray. *Int J Dermatol,* 38: 133-134.
10. **Evereklioglu C**, 2005. Current concepts in the etiology and treatment of Behçet disease. *Surv Ophthalmol,* 50: 297-350.
11. **Forton F, Germaux MA, Brasseur T, De Liever A, Laporte M, Mathys C, Sass U, Stene JJ, Thibaut S, Tytgat M, Seys B,** 2005. Demodicosis and rosacea: epidemiology and significance in daily dermatologic practice. *J Am Acad Dermatol,* 52: 74-87.
12. **Forton F, Seys B,** 1993. Density of *Demodex folliculorum* in rosecea: A case control study using standardized skin-surface biopsy. *Br J Dermatol,* 128: 650-659.
13. **Gao YY, Di Pascuale MA, Li W, Liu DT, Baradaran-Rafii A, Elizondo A, Kawakita T, Raju VK, Tseng SC,** 2005. High prevalence of *Demodex* in eyelashes with cylindrical dandruff. *Invest Ophthalmol Vis Sci,* 46: 3089-3094.
14. **Georgala S, Katoulis AC, Kylafis GD, Koumantaki-Mathioudaki E, Georgala C, Aroni K,** 2001. Increased density of *Demodex folliculorum* and evidence of delayed hypersensitivity reaction in subjects with papulopustular rosacea. *J Eur Acad Dermatol Venereol,* 15: 441-444.
15. **Herron MD, O'reilly MA, Vanderhooft SL**, 2005. Refractory Demodex folliculitis in five children with acute lymphoblastic leukemia. *Pediatr Dermatol*, 22: 407-411.
16. **Ivy SP, Mackall CL, Gore L, Gress RE, Hartley AH,** 1995. Demodicidosis in childhood acute lymphoblastic leukemia; an opportunistic infection occurring with immunosuppression. *J Pediatr,* 127: 751-754.
17. **Jansen T, Kastner U, Kreuter A, Altmeyer P, 2001**. Rosacea-like demodicidosis associated with acquired immunodeficiency syndrome. *Br J Dermatol*, 144: 139-142.
18. **Karincaoğlu Y, Bayram N, Aycan Ö, Esrefoglu M**, 2004. The clinical importance of *Demodex folliculorum* presenting with nonspecific facial signs and symptoms. *J Dermatol,* 31: 618-626.
19. **Karincaoglu Y, Esrefoglu Seyhan M, Bayram N, Aycan O, Taskapan H**, 2005. Incidence of *Demodex folliculorum* in patients with end stage chronic renal failure. *Ren Fail*, 27: 495-499.
20. **Arıcı MK, Sümer Z, Toker Mİ, Erdoğan H, Topalkara A, Akbulut M**, 2005. The Prevalence of *Demodex folliculorum* in blepharitis patients and the normal population. O*phthalmic Epidemiology,* 12: 287-290.
21. **Kheirkhah A, Casas V, Li W, Raju VK, Tseng SC,** 2007. Corneal manifestations of ocular *Demodex* infestation. *Am J Ophthalmol.,* 143: 743-749.
22. **Moravvej H, Dehghan-Mangabadi M, Abbasian MR, Meshkat-Razavi G,** 2007. Association of rosacea with demodicosis. *Arch Iran Med*, 10: 199-203.
23. **Mumcuoglu KY, Akilov OE,** 2005. The role of HLA A2 and Cw2 in the pathogenesis of human demodicosis. *Dermatology,* 210: 109-114.
24. **Okada AA,** 2006. Behçet's disease: general concepts and recent advances. *Curr Opin Ophthalmol*, 17: 551-556.
25. **Patrizi A, Neri I, Chieregato C, Misciali M,** 1997. Demodicidosis in immunocompetent young children: report of eight cases. *Dermatology*, 195: 239-242.
26. **Pecorella I, Ciardi A, Maedsco A, Di Tondo U,** 1999. Histological findings in the eyelids of AIDS patients. *Acta Ophthalmol Scand*, 77: 564-567.
27. **Sakuntabhai A, Timpatanapong P**, 1991. Topical steroid induced chronic demodicidosis. *J Med Assoc Thai*, 74: 116-119.
28. **Seyhan ME, Karincaoğlu Y, Bayram N, Aycan O, Kuku I,** 2004. Density of *Demodex folliculorum* in haematological malignancies. *J Int Med Res,* 32: 411-415.
29. **Roth AM**, 1979. *Demodex folliculorum* in hair follicles of eyelid skin. *Ann Ophthalmol*, 11: 37-40.

30. **Rufli T, Buchner SA,** 1984. T-cell subsets in acne rosecea lesions and the possible role of *Demodex folliculorum*. *Dermatologica,* 169: 1-5.

31. **Rufli T, Mumcuoglu Y**, 1981. The hair follicle mites *Demodex folliculorum* and *Demodex brevis*: biology and medical importance. A review. *Dermatologica*, 162: 1-11.