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Investigation of *Demodex* Prevalence Due to Mask Use After the COVID-19 Pandemic with Cellophane Tape Method

COVID-19 Pandemisi Sonrası Maske Kullanımına Bağlı Demodex Yaygınlığının Selofan Bant Yöntemi ile Araştırılması

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ABSTRACT

Objective: Demodex species in the family Demodicidae are hair follicle scabies agents. *Demodex* species are transmitted from person to person through close contact, shared towels, make-up materials, etc. This study was conducted to obtain data on the relationship between mandatory mask use and demodicosis during the Coronavirus disease-2019 (COVID-19) period.
Methods: The study included 510 students who used masks in necessary environments since the beginning of the pandemic and participated in the study voluntarily. Cellophane tapes were applied to the relevant areas three times. In addition, both eye lashes were pulled from both eyes and adhered to the cellophane tapes. The samples were examined under light microscope at different magnifications (x10, x40).
Results: *Demodex folliculorum* and *Demodex* brevis were detected in 38 (7.5%) of 510 students. *D. folliculorum* was detected in 33 of the positive students and both *D. folliculorum* and *D. brevis* were detected in 5 students, 2 of whom were male and 3 of whom were female. *Demodex* spp. was found in 12 (4.9%) of 245 students who answered "no" to the question "Do you have acne or skin complaints on your face?" and in 26 (9.8%) of 265 students who answered "yes". While this parameter was statistically significant, the other parameters were not statistically significant.
Conclusion: The COVID-19 period has changed people's lifestyles and habits in many ways. It has made the use of masks obligatory. During mandatory mask use, factors that may increase the presence of *Demodex* spp. should not be ignored.
Keywords: COVID-19, *Demodex folliculorum*, *Demodex brevis*, face mask, university students

ÖZ

Amaç: Demodicidae familyasında yer alan *Demodex* türleri kıl folikülü uyuzu etkenleridir. *Demodex* türleri insandan insana yakın temas, ortak kullanılan havlular, makyaj malzemeleri vb. yollarla bulaşır. Bu çalışma, Koronavirüs hastalığı-2019 (COVID-19) sürecinde zorunlu maske kullanımı ile demodikozis arasındaki ilişki hakkında veri elde etmek amacıyla yapılmıştır.
Yöntemler: Çalışmaya pandeminin başlangıcından itibaren zorunlu ortamlarda maske kullanan ve çalışmaya gönüllü olarak katılan 510 öğrenci dahil edilmiştir. Selofan bantlar ilgili bölgelere üç kez uygulanmıştır. Ayrıca her iki göz kirpikleri her iki gözden çekilerek selofan bantlara yapıştırılmıştır. Örnekler ışık mikroskobu altında farklı büyütmelerde (x10, x40) incelenmiştir.
Bulgular: *Demodex folliculorum* ve *Demodex brevis* 510 öğrencinin 38'inde (%7,5) tespit edilmiştir. Pozitif öğrencilerin 33'ünde *D. folliculorum*, 2'si erkek 3'ü kız olmak üzere 5'inde hem *D. folliculorum* hem de *D.brevis* tespit edilmiştir. "Yüzünüzde sivilce veya cilt şikayetiniz var mı?" sorusuna "hayır" cevabı veren 245 öğrencinin 12'sinde (%4,9), "evet" cevabı veren 265 öğrencinin 26'sında (%9,8) *Demodex* spp. bulunmuştur. Bu parametre istatistiksel olarak anlamlı bulunurken, diğer parametreler istatistiksel olarak anlamlı bulunmamıştır.
Sonuç: COVID-19 dönemi insanların yaşam tarzlarını ve alışkanlıklarını birçok yönden değiştirmiştir. Maske kullanımını zorunlu hale getirmiştir. Zorunlu maske kullanımı sırasında *Demodex* spp. varlığını artırabilecek faktörler göz ardı edilmemelidir.
Anahtar Kelimeler: COVID-19, *Demodex folliculorum*, *Demodex* brevis, yüz maskesi, üniversite öğrencileri

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Türkiye PARAZİTOLOJİ Dergisi

INTRODUCTION

Demodex species in the family Demodicidae are "hair follicle scabies agents" that usually settle in facial hair and sebaceous follicles in humans and present with various symptoms. *Demodex folliculorum* is reported to be the most common ectoparasite in humans. *Demodex* species show cosmopolitan distribution and are widely observed all over the world and in our country (1).

Demodex spp. are transmitted from person to person through close contact, shared towels, make-up materials, etc. (1-3). Although it is claimed to be apathogenic by some researchers, it is also known to be pathogenic in cases such as perioral dermatitis, immune system weakness, and the presence of acne vulgaris (1,2). The diagnosis of demodicosis can be made with a standard superficial skin biopsy (SSSB) sample including the follicle contents; by direct slide-to-lamel examination of epilated eyelashes; and by microscopic examination of the material obtained by sticking and pulling the cellophane tape (CT), which is widely used especially in field applications, to the suspicious area (1).

It is recommended that personal care and cleaning products and hand and face towels should not be shared for individual and social protection against infestation. It has been reported that inadequate skin cleansing or keeping the skin moist (such as the use of masks) may create a suitable environment for the parasite to settle (1). With the Coronavirus disease-2019 (COVID-19) pandemic, public health authorities are advising people to wear face masks to reduce respiratory transmission. Wearing a face mask can cause changes in the microenvironment, skin barrier function, and microbiome of human skin. Protective equipment such as masks and gloves used for protection can cause skin diseases such as acne, folliculitis, seborrheic dermatitis, and eczema as a result of inappropriate use (4-6).

This study was planned considering the possibility that prolonged moist skin conditions due to mask use may create a suitable environment for the settlement of parasites of the genus *Demodex* as well as skin disorders. This study was conducted to obtain data on the relationship between the mandatory use of masks and demodicosis. We believe that our study results will contribute to the precautions that can be taken in similar situations that we may encounter in the future.

METHODS

Working Group

This study was conducted with the permission of Sivas Cumhuriyet University Non-Interventional Clinical Research Ethics Committee with the decision of 2021-11/45 dated 17.11.2021. This study was conducted between December 2021 and June 2022. The population of the study consisted of students studying at Sivas Cumhuriyet University Faculty of Medicine, Faculty of Dentistry, Faculty of Pharmacy, Faculty of Health Sciences, Vocational School of Health Services, Faculty of Veterinary Medicine. The age range of the students in the study was 18-23 years old. The study included 510 students who used masks in necessary environments since the beginning of the pandemic and participated in the study voluntarily.

Demodex spp. Examination

After short seminars about *Demodex* spp. and Demodicosis, volunteer students filled out the questionnaire and consent forms. Afterwards, CT samples were taken from the nose, forehead, cheeks, and lower lip. Cellophane tapes were applied to the relevant areas three times each and withdrawn by pressing on them. In addition, both eye lashes were pulled from both eyes and adhered to the CTs. *Demodex* species were identified using literature (1,2,5,7). The samples were examined under light microscope at different magnifications (x10, x40) in the laboratory of Sivas Cumhuriyet University Faculty of Medicine, Department of Parasitology.

Statistical Analysis

For the analysis of the data, descriptive statistical methods, means, standard deviations, medians, frequencies and percentages were used by loading the data into the IBM SPSS 22.0 program. Normality assumption was checked according to Kolmogorov-Smirnov or Shapiro-Wilk test. Parametric tests were used for variables that met the parametric assumption; non-parametric tests were used for variables that did not meet the parametric test assumption. $P<0.05$ was considered statistically significant.

RESULTS

D. folliculorum and/or *D. brevis* were detected in 38 (7.5%) of 510 students (Figures 1, 2). *D. folliculorum* was detected in 33 of the positive students and both *D. folliculorum* and *D. brevis* were detected in 5 students, of which 2 were male and 3 were female.

The distribution of the presence of *Demodex* spp. in the students according to the various questions asked is shown in Table 1. *Demodex* spp. was found in 12 (4.9%) of 245 students who answered "no" to the question "Do you have acne or skin complaints on your face?" and in 26 (9.8%) of 265 students who answered "yes". It was statistically significant ($p=0.035$, $p<0.05$). Other parameters were not statistically significant (Table 1).

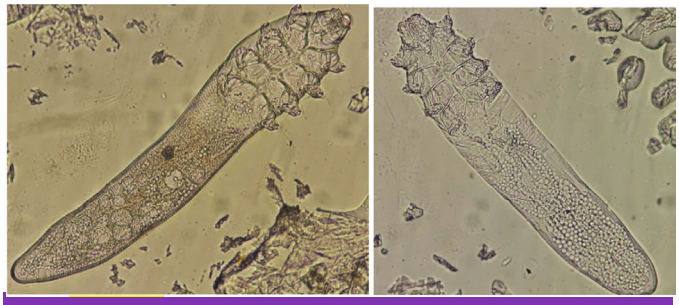

Figure 1. *D. folliculorum* adult (x400)

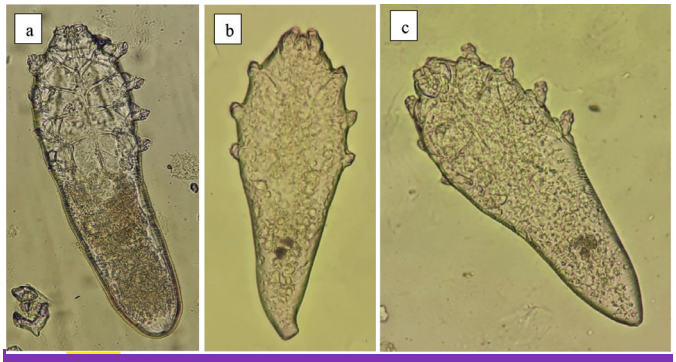

Figure 2. *D. brevis*, a-adult, b-larva, c-nymph (x400)

Table 1. Distribution of *Demodex* spp. according to different characteristics in the survey questions

DISCUSSION

Demodex species can generally be found on the forehead, eyes, nose and around the mouth where sebaceous glands are high; they can also be found on the scalp, outer ear, chest and genital area (2). It can be found all over the world and in all races. The total infestation rate in different study groups generally varies between 17% and 72% in healthy people and can reach up to 100% in people over 96 years of age (1).

It has been reported that *Demodex* spp. can be transmitted more easily through close contact in crowded living spaces such as dormitories, kindergartens, etc., and in the common use of cleaning equipment (7). Some molecular analyses show that frequent and close physical contact leads to mite transmission and haplotypes are likely to be common within families (8).

In university students; Çetinkaya et al. (7), 35.9% in 92 students; Miman et al. (9), 11.0% in 100 students; Kaplan et al. (10), 10.07% in 258 students; Özdemir et al. (11), 47.4% in 270 students; Zeytun et al. (12), 50.1% in 385 students; Sevgen and Mor (13), 42.7% in 375 students; Yilmaz and Akkas (14), 34.5% in 171 students; Ding and Huang (15), 11.5% in 613 students; Isa et al. (16), 17.2% in 390 students were found to have *Demodex* spp.

There is no standard method for *Demodex* examinations; CT method, squeezing method or skin scraping can be used. Liwtin et al. (1) found a 91% positivity rate with the CT method and 34% with the compression method. They recommend the SSSB method as the most commonly used method to compare mite densities between patients with dermatosis and healthy controls (1). However, it has been reported in the literature that the SSSB technique has some limitations (17). Çetinkaya et al. (7) and Miman et al. (9) and, on the other hand, stated that they found the SSSB method more successful and the CT method more unsuccessful. In our study, *Demodex* spp. was detected in 38 (7.5%) of 510 university students who had CT taken from different parts of their faces and answered the survey questions. We believe that the use of only the CT method may be the reason for the low detection rate of *Demodex* spp.

Studies investigating the presence of *Demodex* spp. especially in patients with different dermatologic symptoms on the face have also been conducted. In these studies, Karabay and Çerman (3), 52.0%; Yazısız et al. (18), 69.9%; Maldonado-Gómeza et al. (19), 29.7% *Demodex* spp. were detected and *Demodex* infestation was associated with acne vulgaris, rosacea, and seborrheic dermatitis. Of the people in our study, 163 had COVID-19 and *Demodex* spp. was detected in 14 of them. However, no statistically significant result was found between COVID-19 and *Demodex* spp.

When the relationship between the duration of daily mask wearing, frequency of mask replacement and the presence of *Demodex* spp. was examined, no statistically significant result was found. Some other studies have emphasized that long-term mask use increases the risk of adverse skin reactions, and that

Table 1. Distribution of *Demodex* spp. according to different characteristics in the survey questions

Survey questions		*Demodex* spp.						
		Positive		Negative				
		n	%	n	%	Total	χ^2	p
Gender	Male	17	8.2	191	91.8	208	0.266	0.606
	Female	21	7.0	281	93.0	302		
Have you had a COVID-19 infection?	Yes	14	8.6	149	91.4	163	0.450	0.502
	No	24	6.9	323	93.1	347		
How often did you change masks daily?	1 per day	24	8.3	264	91.7	288	1.831	0.608
	2 per day	5	4.6	103	95.4	108		
	1 per week and >	7	8.6	74	91.4	81		
	Other/irregular	2	6.1	31	93.9	33		
How many hours did you wear your mask per day?	1-3 hours	11	7.0	146	93.0	157	3.277	0.351
	3-6 hours	18	7.8	214	92.2	232		
	6-9 hours	9	9.9	82	90.1	91		
	9 hours and >	0	0.0	30	100.0	30		
What is your daily face washing habit-frequency?	1 per day	11	11.1	88	88.9	99	2.387	0.127
	1 per day >	27	6.6	384	93.4	411		
Do you have acne, skin complaints on your face?	Yes	26	9.8	239	90.2	265	4.437	0.035
	No	12	4.9	233	95.1	245		
Did you use medication for your complaint?	Yes	11	10.0	99	90.0	110	1.321	0.250
	No	27	6.8	373	93.3	400		
Do you use cosmetics (aftershave, moisturizing cream, foundation cream, powder, etc.)?	Yes	30	8.5	322	91.5	352	1.893	0.166
	No	8	5.1	150	94.9	158		
General total		38	7.5	472	92.5	510		

COVID-19: Coronavirus disease-2019

those who do not change their face masks every day are at greater risk of adverse skin reactions than the group that uses masks (19). It has been reported that wearing a mask for several hours daily may cause changes in skin microbiota, and skin barrier functions, and normal skin flora may become pathogenic. It has been stated that demodicosis should be considered in the differential diagnosis in patients with mask-related rash during the COVID-19 pandemic (20,21). It is stated that masks cause microenvironmental changes in the skin through dehydration, sebum and pH increase; *D. folliculorum*, which is considered to be a trigger in rosacea, increases inflammation by taking advantage of sebum overproduction (20,22,23). Avşar et al. (24) investigated *Demodex* spp. frequency, mask use, and personal hygiene habits among medical school students during the COVID-19 pandemic. The researchers found *Demodex* spp. presence at a rate of 47.5% in samples taken from the cheek area and 13.9% in samples taken from the forehead area of those using masks. They commented that the more frequent occurrence in the cheek area may be due to prolonged mask use during the COVID-19 pandemic (24). In a study investigating adverse skin reactions due to the use of protective equipment by healthcare workers during the pandemic process, Turan and Nacar (25) found itching in 23%, scarring on the bridge of the nose in 18%, and pimple/acne formation in the area where the mask was worn in 13% due to mask use.

In studies examining whether the prevalence of *Demodex* spp. infection varies according to gender difference, no statistically significant result was found (2,7,9,11-14,18). In our study, there was no statistically significant result between genders. The fact that it is found slightly higher in men is interpreted as androgen-induced higher sebum production in men and exogenous lipid applications in cosmetics may affect the growth of *Demodex* mites (2). However, Isa et al. (16) found statistically significant results in men compared to women in their study. The researchers attributed this to the fact that men are generally more active throughout the day, which may result in more sebum and sweat secretion (16).

When the relevant literature is examined; results and interpretations differ in terms of the presence of *Demodex* spp. in issues involving personal hygiene. For example, Karaman et al. (26) reported that frequent face washing may cause dryness in the skin, as a result of which the rate of mites will be less on clean skin; Zeytun et al. (12) found that daily face washing frequency and shared towel use affected the frequency of *Demodex* spp. and *Demodex* infestation decreased with increasing number of daily face washes; Forton et al. (27) reported that *Demodex* mites were less common in those who washed their face with soap, and that there was a significant decrease in the frequency of parasites in those who washed their face three times or more daily and had regular skin care. There are also studies in the literature reporting that *Demodex* infestation increases as the frequency of face washing and bathing increases. However, it was also reported that there was no statistically significant result between personal hygiene, self-care and the presence of *Demodex* spp. in these studies (10,13,18,28,29).

In our study, the presence of *Demodex* was found to be 8.5% in those who used cosmetic materials and make-up, and 5.1% in those who did not use it. However, this result was not statistically significant (Table 1). In some other studies, although not statistically significant, it was reported that the presence of *Demodex* was found more frequently in those who did not wear make-up and did not use cosmetics (13,18,24). On the other hand, Okyay et al. (29) reported that living conditions in crowded groups, frequency of daily face washing and hygienic and cosmetic practices such as lotion use did not affect the prevalence of *Demodex* spp. Çetinkaya et al. (7) reported a statistically significant relationship between the use of facial cleansing products only and parasite positivity. They emphasized that skin care and hygiene decreased the frequency of parasites. The researchers interpreted that cosmetics contribute to the destruction of *Demodex* mites by mechanically occluding the follicle, preventing the migration and respiration of the parasite, with a toxic effect because they contain antiseptics such as alcohol (7). In some other studies, Elston and Elston (2) reported that the application of exogenous lipids in cosmetics may affect the growth of *Demodex* mites; Guner et al. (30) reported that changes in skin pH due to cosmetic products lead to disruption of skin barrier function and moisture imbalance, and that the prevalence of sensitive skin increases as the frequency of cosmetic use increases. It should also be taken into account that *Demodex* spp. can survive for long hours in cosmetic materials and this may contribute to its transmission through shared cosmetics (8).

It has been reported that the pathogenicity of *Demodex* mites increases with factors such as neglect and improper skin cleansing, intensive use of cosmetic products, and increased sebum production with sweating (30). It is also reported that *Demodex* spp. may be the cause of chronic inflammatory eruptions of the skin resembling bacterial folliculitis, rosacea, perioral dermatitis and otitis externa (2). Free fatty acids and triglycerides contribute to skin acidity. But although this acidic environment is protective against microorganisms, it is claimed that it has no effect on parasites and even facilitates the presence of parasites (30). Litwin et al. (1) emphasize that higher mite prevalence is observed in rosacea, seborrheic dermatitis, perioral dermatitis, blepharitis and chalazion; the situation is exacerbated when hormonal abnormalities or chronic diseases in the host organism coexist. Nobeyama et al. (31) found *Demodex* in 88.2% of rosacea patients; Avşar et al. (24) found *Demodex* in 52.8% of those with acne and 40.0% of those with other skin problems. In our study, statistically significant presence of *Demodex* spp. was found in people with dermatologic complaints on the face. In some previous similar studies, there were no statistically significant results related to *Demodex* in patients with various skin complaints (3,7,10,11,13,14,16). This is explained by the fact that the cause of facial problems may be multifactorial such as environmental, hormonal, and personal hygiene. However, no significant result was found between the use or non-use of medication for skin complaints.

CONCLUSION

The COVID-19 period has changed people's lifestyles and habits in many ways. It has made the use of masks obligatory. It is also possible that the presence of *Demodex* spp. may be influenced in different directions by the following factors; due to staying at home for a long time during quarantine and not going out, skin cleansing may have been neglected. When coming from outside, extreme care may be taken to clean, due to fear of contracting the COVID-19 virus. In the COVID-19 process, a lot of antiseptic materials were used. Due to the face mask, there has been a decrease in makeup applications and the use of cosmetics. There

was excessive sweating due to the mask. During quarantine, sebum production increased due to excessive consumption of fatty foods such as nuts. Due to staying at home for long periods of time, close contact has increased. However, the relationship between mandatory mask use and infestation by *Demodex* mites has not been studied in detail.

We think that the continuation of many habits acquired during the COVID-19 period and the use of only the CT method during our study may have caused the presence of *Demodex* spp. to be lower compared to previous studies. During mandatory mask use, factors that may increase the presence of *Demodex* spp. should not be ignored.

*Ethics

Ethics Committee Approval: This study was conducted with the permission of Sivas Cumhuriyet University Non-Interventional Clinical Research Ethics Committee with the decision of 2021-11/45 dated 17.11.2021.

Informed Consent: In order to conduct the study, "approved information form" was obtained from the students who agreed to participate in the study in accordance with the letter of Sivas Cumhuriyet University Faculty of Medicine Dean's Office dated 22.02.2022 and numbered 135575; Sivas Cumhuriyet University Rectorate dated 22.03.2022 and numbered 137740.

Footnotes

*Authorship Contributions

Surgical and Medical Practices: A.D.A., B.B.B., Concept: A.D.A., B.B.B., Design: A.D.A., B.B.B., Data Collection or Processing: A.D.A., B.B.B., Analysis or Interpretation: A.D.A., B.B.B., Literature Search: A.D.A., B.B.B., Writing: A.D.A.

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