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# The incidence of *Demodex* species in skin biopsy specimens diagnosed as actinic keratosis and nonmelanoma skin cancer

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**Background:** The most common types of skin cancers include basal cell carcinoma (BCC) and squamous cell carcinoma (SCC), which are grouped as non-melanoma skin cancers. Actinic keratosis (AK) is a precancerous lesion that may develop into SCC. The pilosebaceous follicle mites, *Demodex folliculorum* and *Demodex brevis*, inhabit most commonly and densely certain facial skin areas where BCC and SCC also develops most frequently.

**Objective:** Determine the prevalence of *Demodex* species in skin biopsy specimens diagnosed as SCC, BCC, and AK.

**Method:** Specimens of the patients whose reports were available were studied in terms of *Demodex*. The specimens were stained using Hematoxylin and Eosin, and evaluated for *Demodex* species positivity.

**Results:** There were *Demodex* species in seven (38.9%) out of 18 AK cases, 12 (31.6%) out of 38 SCC cases, and 26 (44.8%) out of 58 BCC cases of this study. The rate of *Demodex* species in patients diagnosed SCC, BCC, and AK was found to be rather high.

**Conclusion:** *Demodex* species should also be evaluated in the follow-up of the treatment of patients in SCC, BCC, and AK group.

**Keywords:** Actinic keratosis, basal cell carcinoma, *Demodex* species, squamous cell carcinoma

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Basal cell carcinoma (BCC) and cutaneous squamous cell carcinoma (SCC) are the most common malignancies in humans [1]. They constitute approximately 95% of non-melanoma skin cancers (NMSCs) [2]. BCC is usually slow growing and rarely metastasizes, but it can cause clinically significant local destruction and disfigurement. The lesion settles on head and neck in 80% of the cases. Subtypes of BCC with different clinical symptoms and distinctive histological appearance and progress have been defined [3].

Actinic keratoses (AK) are frequently seen on dysplastic epidermal lesions, dealt with as SCC

precursor [4]. They frequently settle in upper extremities, head, and neck. The most important agent in their etiopathogenesis is photoaging. They appear clinically in the form of erythematous or dark brown hyperkeratotic papule or plaque [5]. Unless they are treated, they have the potential to invade the dermis, and cause metastasis [4, 5]. Some authors suggest that AK should be acknowledged as a superficial type of SCC [4].

Exposure to sunlight or ultraviolet (UV) from artificial sources is a major risk factor for skin cancer [1]. Moreover, in the etiopathogenesis of the skin tumors, immunosuppression, chronic inflammation, ionized radiation, genodermatosis, mutation in cell cycle regulating genes, and proximal carcinogens are taken into consideration [3, 6, 7].

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The most common permanent ectoparasites include the *Demodex folliculorum*, which lives alone or in groups in the spaces of hair follicles particularly on face and *D. brevis*, which lives alone deep in the sebaceous glands. *Demodex* species can be found in various places in human body including nasolabial region, base of eyelashes, chin, forehead, outer ear canal, nipple, back, penis, and hips [8].

It has been reported that *Demodex* spread between people through close contact, playing a pathogenic role in rosacea, acne vulgaris, perioral dermatitis, seborrheic dermatitis, micropapular-pruritic dermatitis, and blepharitis [9, 10]. Methods used for diagnostic purposes include cellophane tape, skin scraping, punch biopsy and standardized surface skin biopsy (SSSB). The mite intensity of the parasite per cm<sup>2</sup> is required to be known to detect the pathogenesis of the parasite. Especially, the SSSB method is effective in the diagnosis of the *Demodex* species. Based on this method, the follicular content is collected completely together with the surface part of the stratum corneum where the parasite inhabits, thus making it easier to detect the mite intensity/cm<sup>2</sup> [10].

In this study, we investigated the prevalence of *Demodex* species in skin biopsy specimens diagnosed as SCC, BCC, and AK.

### Material and methods

The ethical council report was granted prior to the study from Medical Faculty of Inonu University. The specimens examined in this study included the skin biopsies brought to the Pathology Laboratory in the Medical Faculty of Inonu University between 2002 and 2005. A retrospective study was conducted on specimens obtained from patients whose reports were released were also examined for *Demodex*. The experiment group of the study comprised the patients whose skin biopsies were examined and diagnosed as SCC, BCC, and AK. All of the archived specimens obtained from the facial skin biopsies of the patients diagnosed SCC, BCC and AK were considered for evaluation.

The specimens were stained using Hematoxylin and Eosin (HE) method, and evaluated for *Demodex* species positivity. The study population consisting of

114 samples was examined by a parasitologist and a pathologist. The both researchers examined the samples in different times, and they obtained similar results. Preparation works for histopathological examination and HE staining did not prevent the incidence of *Demodex* species. No discrimination was done in relation to *D. folliculorum* and *D. brevis*. In the examination of the samples, a *Demodex* species was considered positive. In this retrospective study, we did not take sample from the patients with SSSB, and did not assess the intensity of flow. Specimens were also examined for the percentage of *Demodex* prevalence, according to gender, age and the location of biopsy.

### Statistical analysis

The data were presented in terms of mean, standard deviation or number, and percentages. Normality was proved by Shapiro-Wilk test. Independent samples *t* test, Pearson Chi-square, and Yates' adjusted Chi-square tests were used for the statistical analyses. Analyses were done using SPSS 13.0 software program. A value of  $p < 0.05$  was considered as statistically significant.

### Results

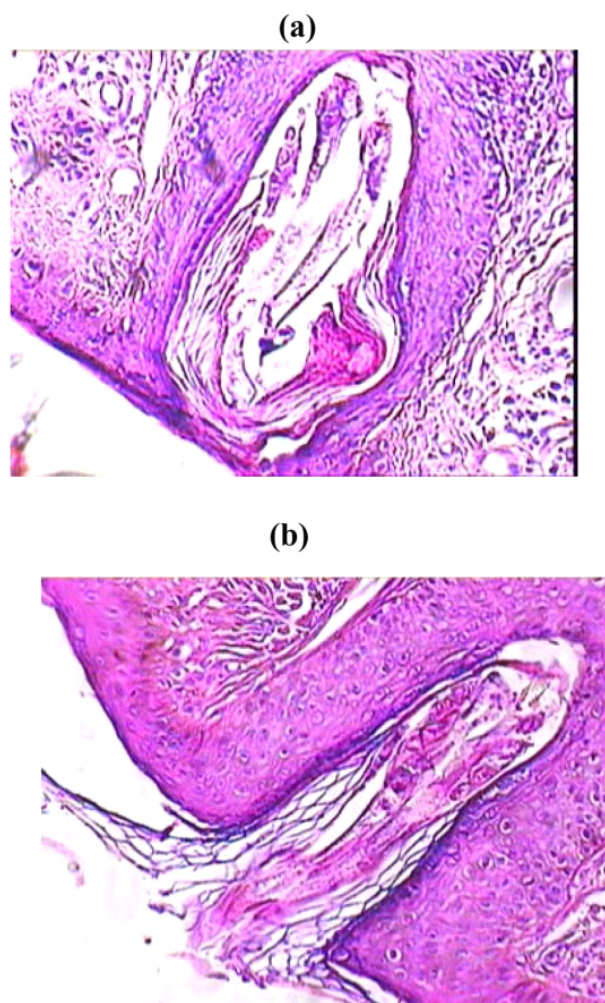
The analysis revealed the prevalence of *Demodex* species in 38.9% of AK, 39.6% of SCC, and 44.8% of BCC cases (see **Fig. 1**).

**Table 1** shows the distribution of the findings about *Demodex* species detected according to diagnosis.

The distribution of *Demodex* species, according to gender variable is shown in **Table 2**. No significant difference was observed between the presence of *Demodex* species and gender variable ( $p=0.125$ ).

The distribution of the prevalence of *Demodex* species according to the location of biopsy is shown in **Table 3**. There is no significant difference between the presence of *Demodex* species and location of biopsy ( $p=0.21$ ).

The distribution regarding the prevalence of *Demodex* species according to age groups is shown in **Table 4**. There is no significant difference between presence and absence of *Demodex* species in terms of age variable ( $p=0.63$ ).



**Fig. 1** *Demodex* species in hair follicles in biopsy specimens. (a) x100 (HE staining), (b) x200 (HE staining).

**Table 1.** Distribution (%) of the *Demodex* species according to diagnosis.

Diagnosis	Positive Amount (%)	Negative Amount (%)	Total Amount (%)
AK	7 (8.9)	11 (61.1)	18 (100)
SCC	12 (31.6)	26 (8.4)	38 (100)
BCC	26 (44.8)	32 (55.2)	58 (100)

**Table 2.** The presence of *Demodex* species according to gender.

Gender	<i>Demodex</i> species		Total Amount (%)
	No Amount (%)	Yes Amount (%)	
Male	39 (68.0)	18 (32.0)	57 (100.0)
Female	30 (53.0)	27 (47.0)	57 (100.0)
<b>Total</b>	69 (61.0)	45 (39.0)	114 (100.0)

**Table 3.** The presence of *Demodex* species according to location of biopsy was obtained.

Location	<i>Demodex</i> species		
	No Amount (%)	Yes Amount (%)	Total Amount (%)
Forehead	8 (80.0)	2 (20.0)	10 (100.0)
Nose	23 (54.8)	19 (45.2)	42 (100.0)
Chin	2 (66.7)	1 (33.3)	3 (100.0)
Hairy skin	6 (75.0)	2 (25.0)	8 (100.0)
Cheek	17 (48.6)	18 (51.4)	35 (100.0)
Eyebrow	10 (83.4)	2 (16.6)	12 (100.0)
Ear	3 (75.0)	1 (25.0)	4 (100.0)
<b>Total</b>	<b>69 (61.0)</b>	<b>45 (39.0)</b>	<b>114 (100.0)</b>

**Table 4.** The distribution of presence of *Demodex* species and age groups.

<i>Demodex</i>	Number	Mean	Standard deviation	P-value
Yes	69	63.3	1.8	0.63
No	45	64.4	1.7	

## Discussion

Basal cell carcinoma is the most common malignancy in humans. Its incidence continues to rise [11]. SCC is the second most common skin cancer after BCC [12]. Most skin cancers are settled on head and neck. AK are precancerous formations, mostly seen in upper extremities, head, and neck. While exposure to the UV radiation in sunlight is major predisposition in skin cancer, other factors play a role in their etiology [13].

There are two species of *Demodex* parasites in humans, *D. folliculorum* and *D. brevis*. There are different views about the pathology and clinical symptoms caused by these mites in humans [14, 15]. Some researchers consider inhabitation of *Demodex spp* in pilosebaceous follicles harmless, and others reported that *D. folliculorum* can play an etiopathogenic role in rosacea, acne vulgaris, blepharitis, perioral dermatitis, pustular folliculitis, papular-pustular lesions on hairy skin, and pustular lesions in acquired immune deficiency syndrome [8].

It was reported that the prevalence of *Demodex* species increases as the patients grow older [16]. Baysal et al. [9] evaluated the association between presence of *Demodex* species and different age groups, and found the parasite in 8.3% of the patients among 11-15 age group and 12.7% among 16-20 age group. In another study, the prevalence of *Demodex*

was detected as 20% among  $\leq 20$  age group and 53.5% among  $21 \leq$  age group [17]. Similarly in this study, 45 out of 114 patients aged 63.71 on average were detected *Demodex* positive.

Roihu et al. [18] reported the prevalence of *Demodex* as 59% among male and 30% among female. Baysal et al. [9] reported *Demodex* positivity among 11.9% of the 67 female with acne vulgaris and 11.7% of the 34 male subjects. In the present study 18 out of 57 male patients and 27 out of 57 female patients were found to have the parasite. The prevalence of the parasite between male and female differs in different studies. This can be accounted for with other mediating factors such as the age of the subjects, the place they live, their life styles, and researchers.

In a previous study, parasite was detected among 26% of the health personnel working in the autopsy room [19]. Similarly in another study, 37.7% out of 75 subjects diagnosed blepharitis and 32% out of the 125 control subjects were found to have the parasite [20]. In the present study, *Demodex* species were observed in 38.9% of 18 AK, 39.6% of 38 SCC, and 44.8% out of 58 BCC cases.

The pathogenesis of *D. folliculorum* is not exactly known. It has been reported that the immunologic reactions developed against the parasite and the proliferation of *D. folliculorum* due to immunological

defects play a role in emergence of skin lesions [21].

*D. folliculorum* has been also detected in immune compromised children such as with acute lymphoblastic leukemia. It was reported that this parasite caused rosacea and perioral dermatitis in these children [22]. It was reported that though *Demodicidosis* is seen less among children, it was reported to be more common among AIDS and lymphoproliferative disorders [23]. In another study, 12.76% out of 47 patients with chronic renal failure were found to have the parasite [10].

Sun et al. [24] investigated the *D. folliculorum* infestation in facial epidermal neoplasms. They found the highest levels in BCC (56%), while low levels of infestation were found in seborrheic keratosis, trichilemmoma, and SCC. In the present study, *Demodex* species were also observed among patients with BCC, SCC, and AK to a high extent.

In conclusion, *Demodex* species should be considered in the follow-up of the treatment of patients in SCC, BCC, and AK group.

#### Acknowledgement

We thank Prof. Engin N. Aydin, Prof. Nilgun Daldal, Assist. Prof. Nese Karadag, and Assist. Prof. Metin Atambay for their contributions to this study. The authors have no conflict of interest to declare.

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