

Case report: Isolation of Scopulariopsis brevicaulis from Wistar Rats

Özlem Şahan Yapıcıer¹* ⓑ, Mehmet Kaya² ⓑ, Zeki Erol³ ⓑ, Dilek Öztürk⁴ ⓑ

^{1,24} Faculty of Veterinary Medicine, Mehmet Akif Ersoy University, Department of Microbiology, Burdur, TURKEY ³ Mehmet Akif Ersoy University, Experimental Animal Production and Experimental Research Center, Burdur, TURKEY

Geliş Tarihi / Received: 13.07.2020, Kabul tarihi / Accepted: 07.12.2020

Abstract: *Scopulariopsis brevicaulis* is a saprophytic fungus that has wide geographic distribution. This study describes a case of hair loss and skin lesions observed in male and female Wistar rats due to *Scopulariopsis brevicaulis* infection in Turkey. Skin scrapings and hair samples from three male and two female rats were provided by the Experimental Animal Production and Experimental Research Center of Mehmet Akif Ersoy University to the Faculty of Veterinary Medicine, Department of Microbiology Laboratory in Burdur for analysis in July 2019. Microbiological methods were used for species identification and *Scopulariopsis brevicaulis* was isolated from all of the samples. The rats completely recovered without treatment and had no recurrence of clinical signs at one month post-sampling. This study is the first report of *S. brevicaulis* causing an infection in Wistar rats in Turkey.

Keywords: Laboratory animals, mycological examination, rats, saprophyte, Scopulariopsis sp

Olgu sunumu: Wistar Ratlarından Scopulariopsis brevicularis izolasyonu

Özet: *Scopulariopsis brevicaulis*, geniş coğrafi dağılımı olan saprofitik bir mantardır. Bu olgu, Türkiye'deki erkek ve dişi Wistar ratlarında *Scopulariopsis brevicaulis* infeksiyonuna bağlı olarak gözlenen tüy kaybı ve deri lezyonlarını tanımlamaktadır. Üç erkek ve iki dişi albino Wistar ratından deri kazıntıları ve tüy örnekleri, Temmuz 2019'da analiz için Mehmet Akif Ersoy Üniversitesi Deney Hayvanları Üretim ve Deneysel Araştırma Merkezi tarafından Burdur Veteriner Fakültesi Mikrobiyoloji Anabilim Dalı Laboratuvarına gönderildi. Mikrobiyolojik yöntemler tür identifikasyonu için kullanıldı ve tüm örneklerden *Scopulariopsis brevicaulis* izole edildi. Tüm ratlar tedavi gerektirmeden iyileşti ve örneklemeden bir ay sonra klinik belirtiler nüksetmedi. Bu çalışma, *S. brevicaulis*'in Türkiye'deki Wistar ratlarında infeksiyona neden olduğunu bildiren ilk rapordur.

Anahtar kelimeler: Laboratuvar hayvanları, mikolojik muayene, ratlar, saprofit, Scopulariopsis sp

Introduction

Scopulariopsis (Microascus) spp. belongs to the ascomycete family Microascaceae and is a ubiquitous, saprobic mold found worldwide in soil, plant debris, wood, paper, animal matter, air, and moist indoor environments (Abbott et al. 1998; Anandan et al. 2008). The genus Scopulariopsis, and predominantly S.brevicaulis, is known to be an opportunistic pathogen that is thought to cause invasive (immunocompetent and immunosuppressed patients) and noninvasive infections such as, onychomycosis, keratitis, conjunctivitis, endocarditis, and disseminated in animals and humans (Ragge et al. 1990; Tosti et al. 1996; Iwen et al. 2012; Cawcutt et al. 2015; Kordalewska et al. 2016; Hampson et al. 2019).

This study aimed to describe a case of hair loss and skin lesions due to *Scopulariopsis brevicaulis* infection in male and female Wistar rats in Turkey.

Material and Methods

A total of three male and two female rats were housed in cages with controlled 8 h of light and 16 h of darkness daily. The environmental temperature and humidity were maintained at $21\pm1^{\circ}$ C and 70%, respectively. Rats were given *ad libitum* access to food and water.

Two weeks before analysis, progressive alopecia was observed in five-week-old Wistar albino rats at the Experimental Animal Production and Experimental Research Center of Mehmet Akif Ersoy University. Upon physical examination, alopecia was observed across the entire bodies of males and females without additional signs of disease. There was no previous history of alopecia. Skin scrapings and hair samples were collected in sterile sputum cups (FIRATMED, Turkey) from the various lesions on the bodies of three males and two females and immediately submitted to the Faculty of Veterinary Medicine, Department of Microbiology Laboratory in Burdur in July 2019 for analysis as shown in Figures 1 and 2.

Yazışma adresi / Correspondence: Özlem Şahan Yapıcıer, Faculty of Veterinary Medicine, Mehmet Akif Ersoy University, Department of Microbiology Istiklal Campus, 15030, Burdur, Turkey, Phone: +90 248 2132064, +90 532 5849864, E-posta: ozlem-sahan@hotmail.com, ozlemsahan@mehmetakif.edu.tr

ORCID IDs of the authors: 10000-0003-3579-9425 • 20000-0002-7927-4885 • 30000-0002-1563-0043 • 40000-0002-9643-8570



Figure 1. S. brevicaulis lesions on the male rats.



Figure 2. S. brevicaulis lesions on the female rats.

Isolation and Identification

For microbiological examination, part of the samples was processed in 10-20% potassium hydroxide (KOH) for 30 min and then examined by light microscopy using 400x magnification. The remaining samples were seeded on Sabouraud Dextrose agar (SDA, OXOID) plates supplemented with chloramphenicol (0.05 mg/mL) and separately incubated at 25°C and 37°C for 10 days. Plates were examined on a daily basis as previously described and isolated colonies were subsequently stained with lactophenol blue (Larone 1995). The isolated fungal colonies were stained with lactophenol blue and identified as *S. brevicularis* based on macroscopic and microscopic characteristics.

Results and Discussion

Breeding colonies of laboratory animals are of utmost importance in research for investigating a wide range of biomedical topics, such as understanding mechanisms of disease, developing disease prevention or treatment options, and evaluating health risks present in our living environment (Nicklas 2004). Although breeding conditions (e.g., diet, light conditions, bedding) for laboratory animals continue to improve, various infectious agents remain prevalent among colonies, especially fungal species (Park et al. 2006).

In this study, KOH preparations of skin scrapings and hair samples showed no hyphae. Identification of *S. brevicaulis* was confirmed by moderately fast growth at 25°C and poor growth at 37°C (Figure 3) with characteristic colonies' surface at first white, powdery and glabrous, and then becomes powdery light brown with a light tan periphery, and reverse was tan with the brownish center. Overall, microscopically showed hyaline or dematiaceous septate hyphae with annelids (brush-like groups form) that formed on a branched penicillin-bearing lemonshaped conidia are shown in Figures 4 and 5.

There are limited data of clinical and experimental treatment of *S. brevicaulis* infections for animals and humans, and even studies were reported that appropriate therapy for *Scopulariopsis* infections has yet to be defined (Aguilar et al. 1999; Cuenca-Estrella et al. 2006; Ozturk et al. 2009; Miossec et al. 2011). Despite the high antifungal resistance of *S. brevicularis*, researchers recommend all common antifungal agents such as amphotericin B (AMB) and voriconazole (VRC) (Cuenca-Estrella et al. 2006; Skóra et al. 2015). In this study, rats completely recovered without treatment and had no recurrence of clinical signs at one month post-sampling. It was thought that the cause of the total recovery of rats without any use of antifungal agent from opportunistic *S. brevicularis* infection was due to have no immune problems.



Figure 3. Macroscopic morphology of *S. brevicaulis* on SDA agar after 10 days at 37°C.



Figure 4. Macroscopic morphology of *S. brevicaulis* on SDA agar after 10 days at 25°C (A and B images).



Figure 5. Branching and septated hyphae with conidiophores and conidia in chains seen on lactophenol cotton blue stain (A, B, and C images).

Scopulariopsis brevicularis cause invasive and non-invasive infections in animals and humans and unfortunately, there are no recommended therapies in previous researches (Bonifaz et al. 2007; Issakainen et al. 2007). The improper diagnostic methods and the high level of antifungal resistance (Aguilar et al. 1999; Sandoval et al. 2013; Skóra et al. 2015) may be responsible for the high mortality of Scopulariopsis infections (Issakainen et al. 2007). This case is the first to describe S. brevicaulis infection in Wistar rats from Turkey. Interestingly, S. brevicaulis was reported as an etiological agent of skin infections in two goats by Ozturk et al. (2009) in the same province. There have also been several previous reports of a high incidence of S. brevicaulis in humans and animals (dogs, cats, and large animals), which was unexpected (Romano et al. 2005; Costa et al. 2019; Sri-Jayantha et al. 2019). Moreover, the detection of this fungal agent in skin infections might be correlated with animal matter, such as hair, skin scales, sweat, and other organic material on the ground or wood shavings of cages. Mantovani et al. (1982) described this phenomenon as "animalization". Animal age, sex, and housing climate were found to be factors in previously described S. brevicaulis infections (Couto et al. 2014); however, these were not significant factors in this study.

Microbiological detection methods have been widely used to diagnose *Scopulariopsis* at the genus level, but maybe difficult for identifying species (Yang et al. 2012). Thus, distinctive features of *Scopulariopsis*, such as colony color and annellidic conidiogenesis, might be useful for identification (Samson et al. 2010).

This is the first report of *S. brevicaulis* infection in Wistar laboratory rats in Turkey in this study. This case may provide a guide for clinicians and microbiologists to diagnose unexpected fungal infections of laboratory animals. Additionally, ensuring strict control of hygiene for providing welfare to laboratory animals can help guarantee that they are protected well against infections.

Ethical approval and consent to participate:

Not applicable.

References

- Abbott SP, Sigler L, Currah RS. (1998) *Microascus brevicaulis* sp. nov., the teleomorph of *Scopulariopsis brevicaulis*, supports placement of *Scopulariopsis* with the Microascaceae. *Mycologia*. 90(2), 297-302. doi: 10.1080/00275514.1998.12026910
- Anandan V, Nayak V, Sundaram S, Srikanth P. (2008) An association of *Alternaria alternata* and *Scopulariopsis brevicaulis* in

cutaneous phaeohyphomycosis. *Indian J. Dermatol Venereol Leprol.* 74(3), 244-247. doi: 10.4103/0378-6323.41371

- Aguilar C, Pujol I, Guarro J. (1999) In vitro antifungal susceptibilities of Scopulariopsis isolates. *Antimicrob Agents Chemother*. 43(6), 1520-1522.
- Bonifaz A, Cruz-Aguilar P, Ponce RM. (2007) Onychomycosis by molds. Report of 78 cases. *Eur J Dermatol.* 17(1), 70 -72. doi: 10.1684/ejd.2007.0092
- Cawcutt K, Baddour LM, Burgess M. (2015) A case of Scopulariopsis brevicaulis endocarditis with mycotic aneurysm in an immunocompetent host. Case Rep Med. 2015871-872. doi. org/10.1155/2015/872871
- Costa FVA, Spanamberg A, Araujo R, Werner J, Ferreiro L. (2019) Feline sino-orbital fungal infection caused by *Aspergillus* and *Scopulariopsis. Acta Sci Vet.* 47(Suppl 1), 383-341. doi: 10.22456/1679-9216.91581
- Couto MS, Pantoja LDM, Mourao CL, Paixao GC. (2014) Fungal microbiota of the hair coat of laboratory animals. *Rev Bras Vet.* 17(1), 52-54.doi: 10.4322/rbcv.2014.143
- Cuenca-Estrella M, Gomez-Lopez A, Buitrago MJ, Mellado E, Garcia-Effron G, Rodriguez-Tudela JL. (2006) *In vitro* activities of 10 combinations of antifungal agents against the multiresistant pathogen *Scopulariopsis brevicaulis*. *Antimicrob Agents Chemother*. 50, 2248-2250. doi: 10.1128/AAC.00162-06
- Hampson ECGM, Gibson JS, Barot M, Shapter FM, Greer RM. (2019) Identification of bacteria and fungi sampled from the conjunctival surface of normal horses in South-East Queensland, Australia. *Vet Ophthalmol.* 22(3), 265-275. doi: 10.1111/vop.12587
- Issakainen J, Heikkilä H, Vainio E, Koukila-Kähkölä P, Castren M, Liimatainen O, Ojanen T, Koskela M, Meurman O. (2007) Occurrence of Scopulariopsis and Scedosporium in nails and keratinous skin. A 5-year retrospective multi-center study. Med Mycol. 45(3):201-209. doi: 10.1080/13693780601103080.
- Iwen P, Schutte SD, Florescu DF, Noel-Hurst RK, Sigler L. (2012) Invasive Scopulariopsis brevicaulis infection in an immunocompromised patient and review of prior cases caused by Scopulariopsis and Microascus species. J Mycol Med. 50(6), 561-569.doi: 10.3109/13693786.2012.675629
- Kordalewska M, Jagielski T, Brillowska-Dabrowska A. (2016) Rapid assays for specific detection of fungiof Scopulariopsis and microascus genera and *Scopulariopsis brevicaulis* species. *Mycopathologia*. 181(7-8), 465-474. doi: 10.1007/s11046-016-0008-5
- Larone DH. (1995) *Medically important fungi: A guide to identification*. Fourth edition, American Society for Microbiology, USA, p. 272-274.
- Mantovani A, Morganti L, Batelli G, Mantovani A, Poqlayen G, Tampieri MP, Vecchi G. (1982) The role of wild animals in the ecology of dermatophytes and related fungi. *Folia Parasitol*. 29(3), 279-284.
- Miossec C, Morio F, Lepoivre T, Le Pape P, Garcia-Hermoso D, Gay-Andrieu F, Haloun A, Treilhaud M, Leclair F, Miegeville M. (2011) Fatal invasive infection with fungemia due to *Microascus cirrosus* after heart and lung transplantation in a patient with cystic fibrosis. J Clin Microbiol. 49(7), 2743-2747. doi: 10.1128/JCM.00127-11
- Nicklas W. (2004) Infectious in laboratory animals: Importance and control. Kaliste E. eds. The Welfare of Laboratory Animals. Kluwer Academic Publishers, Dordrecht, Netherland. p. 153.
- Ozturk D, Adanır R, Turutoglu H. (2009) Superficial skin infectionwith *Scopulariopsis brevicaulis* in two goats. a case report. Bull Vet Inst Pulawy. 53, 361-363.

- Park J-H, Seok S, Baek M, Lee H, Kim D, Cho J, Kim C, Hwang D. (2006) Microbiological monitoring of guinea pigs reared conventionally at two breeding facilities in Korea. *Experimental Animal.* 55(5), 427-432. doi:10.1538/expanim.55.427
- Ragge N, Hart J, Easty D, Tyers A. (1990) A case of fungal keratitis caused by *Scopulariopsis brevicaulis* treatment with antifungal agent and penetrating keratoplasty. Br J Opthalmol. 74(9), 561-562. doi: 10.1128/jcm.00127-11
- Romano C, Gianni C, Difonzo EM. (2005) Retrospective study of onychomycosis in Italy: 1985-2000. *Mycoses*. 48: 42-44. doi: 10.1111/j.1439-0507.2004.01066.x
- Samson RA, Houbraken J, Thrane U, Frisvad JC. (2010) *Food and indoor fungi*. CBS Laboratory Manual Series 2. CBS-Fungal Biodiversity Centre, Utrecht.
- Sandoval-Denis M, Sutton DA, Fothergill AW, Cano-Lira J, Gené J, Decock CA, de Hoog GS, Guarro J. (2013) *Scopulariopsis*, a poorly known opportunistic fungus: spectrum of species in

clinical samples and *in vitro* responses to antifungal drugs. *J Clin Microbiol.* 51(12), 3937-3943. doi: 10.1128/JM.01927-13

- Skóra M, Bulanda M, Jagielski T. (2015) *In vitro* activities of a wide panel of antifungal drugs against various *Scopulariopsis* and *Microascus* species. *Antimicrob Agents Chemother*. 59, 5827-5829. doi: 10.1128 /AAC.00978-15
- Sri-Jayantha L, Matthews KG, Scharf V. (2019) Scopulariopsis brevicaulis rhinosinal infection in a dog. J Am Anim Hosp Assoc. 55: e55102. doi: 10.5326/jaaha-ms-6869
- Tosti A, Piraccini BM, Stinchi C, Lorenzi S. (1996) Onychomycosis due to *Scopulariopsis brevicaulis:* clinical features and response to systemic antifungals. Br J Dermatol. 135(5), 799-802. doi: 10.1111/j.1365-2133.1996.tb03895.x
- Yang Q, Wei J, Chen Z. (2012) Fatal bronchial invasion of Scopulariopsis brevicaulis in an acute monocytic leukemia patient. Diag Microbiol Infect Dis. 73(4), 369-371. doi: 10.1016/j.diagmicrobio. 2012.04.010