Mycopathologia (2017) 182:371–377
DOI 10.1007/s11046-016-0083-7

Incidence of Keratinophilic Fungi from the Selected Soils of Kaziranga National Park, Assam (India)

Sunil Kumar Deshmukh · Shilpa Amit Verekar · Yashwant G. Chavan

Received: 25 June 2016 / Accepted: 24 October 2016 / Published online: 31 October 2016

Abstract Seventy-eight soil samples were collected from the various locations in the vicinity of Kaziranga National Park (Assam), India, during April to October 2009 and screened for the presence of keratinophilic fungi using the hair baiting techniques for isolation. Thirty-nine isolates were recovered and identified by recognition of their macro- and micromorphological features. Their identification was also confirmed by the BLAST search of sequences of the ITS1-5.8S-ITS2 rDNA region against the NCBI/GenBank data and compared with deposited sequences for identification purpose. Eleven species related to seven genera were recorded viz. *Aphanoascus durus* (1.28%), *Arthroderma tuberculatum* (3.84%), *Arthroderma corniculatum* (1.28%), *Chrysosporium indicum* (16.66%), *C. tropicum* (3.84%), *Ctenomyces serratus* (5.12%), *Keratinophyton punsolae* (1.28%), *Microsporum appendiculatum* (1.28%), *Microsporum gypseum* complex (11.53%), *Trichophyton mentagrophytes* (11.28%) and *T. terrestre* (2.56%).

Keywords Kaziranga National Park · Assam · Soil fungi · Keratinophilic fungi · India

S. K. Deshmukh (✉)
TERI-Deakin Nano Biotechnology Centre, The Energy and Resources Institute, Darbari Seth Block, IHC Complex, Lodhi Road, New Delhi 110 003, India
e-mail: sunil.deshmukh@teri.res.in

S. A. Verekar
Department of Microbiology, St. Xavier's College – Autonomous, 5, Mahapalika Marg, Mumbai, Maharashtra 400 001, India
e-mail: shilfa1@rediffmail.com

Y. G. Chavan
Dr. D. Y. Patil Biotechnology and Bioinformatics Institute, Tathwade, Dr. D. Y. Patil University, Pimpri, Pune 411018, India
e-mail: yashwant.chavan@geneombiotechnologies.com

Introduction

Keratinophilic fungi are an ecologically significant group of fungi that decompose keratin—animal-origin protein—which is one of the most abundant and highly stable proteins on earth. They utilize it as a nutrient substrate for growth. The distribution of these fungi depends on different factors, one of which, of vital importance, is human and/or animal presence [1]. Vanbreuseghem reported for the first time the presence of dermatophytes in soil [2] using the hair bait technique. Since then, researchers across the globe are using hair bait technique for the isolation of keratinophilic fungi from soil [3–8]. Some of these fungi are well-known dermatophytes and are known to cause superficial cutaneous infections (dermatophytoses) of keratinized tissues (skin, hair and nails) of humans and animals.

Kaziranga National Park (latitudes 26°30′N and 26°45′N and longitudes 93°08′E to 93°36′E) is one of

the India's finest parks and is situated in Assam, home to several endangered species. Its tropical climate and geographic diversity make it a potentially interesting area to study the distribution of keratinophilic fungi. We, therefore, undertook this study and reported the results obtained.

Materials and Methods

Collection and Processing of Soil Samples

Seventy-eight soil samples were collected from the various localities in the vicinity of Kaziranga National Park (Assam), India, during April to October 2009 (Table 1). The samples were taken from the elevated regions known as chapories, which provide retreats and shelter for animals during floods, soil from water coarse bank, forest soil, grassland soil, barren land soil, the raw road (pagdandi) soil and dropped off feathers. Soil samples were collected by removing a layer of soil not exceeding 3–5 cm in depth with a sterile stainless steel spatula and transferred in sterile polyethylene bags, brought to the laboratory and stored at 15 °C if not processed promptly. Keratinophilic fungi were isolated by the baiting technique of Vanbreuseghem [2] using human hair as keratin bait. For this, sterile Petri dishes, half filled

Table 1 Distribution of fungi in soil samples of Kaziranga National Park

Source of soil samples	Resting areas of animals	Water coarse bank	Forest soil	Grass land soil	Barren land	The raw road (Pagdandi)	Dropped off feathers	Total	Distribution (%)
No. of samples examined	10	10	14	12	10	10	12	78	
No. of samples positive	6	4	7	6	4	6	6	39	
Distribution (%)	60	40	50	50	40	60	50	50	
Aphanoascus durus	–	–	1	–	–	–	–	1	1.28
Arthroderma tuberculatum	–	1	1	–	–	–	1	3	3.84
Arthroderma corniculatum	–	–	–	1	–	–	–	1	1.28
Chrysosporium indicum	1	1	–	3	2	3	3	13	16.66
Chrysosporium tropicum	–	1	1			1	–	3	3.84
Ctenomyces serratus	1		1		–	1	1	4	5.12
Keratinophyton punsolae	–	–		1	–	–	–	1	1.28
Microsporum appendiculatum	1	–	–	–	–	–	–	1	1.28
Microsporum gypseum complex	1	1	2	1	2	1	1	9	11.53
Trichophyton mentagrophytes	1	–		–	–	–	–	1	1.28
Trichophyton terrestre	1	–	1	–	–	–	–	2	2.56
Total	6	4	7	6	4	6	6	39	50.00

with the soil samples and moistened with sterile tap water, were baited by burying sterile human hairs in the soil. These dishes were incubated at room temperature (25 ± 1 °C) and examined daily from the fifth day for fungal growth over a period of 4 weeks.

Isolation and Identification of Keratinophilic Fungi

The growth on hairs was observed under a stereoscopic binocular microscope and was subcultured on SDA (Sabouraud's dextrose agar) plates amended with chloramphenicol (50 mg/l) and cycloheximide (500 mg/l). The plates were incubated for 5 to 10 days at room temperature. The isolated cultures were checked for purity and subcultured to obtain pure cultures. These fungi were identified based on the various available monographs (Sigler and Carmichael [9] Oorschot [10] Currah [11] Arx [12] Cano and Guarro [13]).

Molecular Identification of Keratinophilic Fungi

The DNA sequences of ITS1-5.8S-ITS2 region were used for molecular characterization of the cultures. Fungal genomic DNA was extracted [14]. ITS1 and ITS4 were used as the forward and reverse primers for amplification of ITS1-5.8S-ITS2 rDNA [15]. PCR amplification was performed in 100 µl reaction volumes containing 10X buffer 10 µl, $MgCl_2$ (25 mM) 2 µl, dNTP (10 mM) 2 µl, ITS1 primer (20 pm) 2 µl, ITS4 primer (20 pm) 2 µl, Taq Polymerase (2.5U) 1 µl, DNA Sample (5 µg/ml) 3 µl and Milli Q water 78 µl. The PCR was carried out using a Thermal Cycler (M.J. research, PTC 200) with following conditions: denaturation for 5 min at 94 °C, 34 cycles of (30 s at 94 °C, 30 s at 55 °C, 1 min at 72 °C) extension for 4 min at 72 °C and storage at 4 °C. Controls were used in each set of reactions. The final products were analyzed by agarose gel electrophoresis (Sigma). The PCR products were sequenced using ITS1 and ITS4 primers at Geneombio Technologies Pvt Ltd, Pune, India, using Applied Biosystems 3730 DNA analyzer.

Phylogenetic Analysis

Similarity analysis of the nucleotides was performed by BLAST searches against sequences available in GenBank [16]. For phylogenetic tree construction, multiple sequences were obtained from GenBank and the alignments were performed using MEGA6 [17].

Results and Discussion

Data on the distribution of keratinophilic fungi in the soils of Kaziranga National Park (Assam) are given in Table 1. The data show that only 39 of the 78 samples yielded keratinophilic fungi. A total of eleven species from seven genera were isolated: *Aphanoascus durus* (1.28%), *Arthroderma tuberculatum* (3.84%), *Arthroderma corniculatum* (1.28%), *Chrysosporium indicum* (16.66%), *C. tropicum* (3.84%), *Ctenomyces serratus* (5.12%), *Keratinophyton punsolae* (1.28%), *Microsporum appendiculatum* (1.28%), *Microsporum gypseum* complex (11.53%), *Trichophyton mentagrophytes* (1.28%) and *Trichophyton terrestre* (2.56%).

All the eleven strains yielded unique PCR amplicon. The amplicon size of the ITS1-5.8S-ITS2 rDNA regions for the eleven strains was ranging from 409 to 571 bp, where *C. indicum* is smallest and *T. terrestre* is largest in size. The other species showed an amplicon size of approximately 600 bp. The sequences were compared with sequences deposited in the NCBI database for taxonomic identification, and a phylogenetic tree was constructed with the closely related type strain sequences based on rRNA gene sequences (ITS region) using the maximum composite likelihood method. The phylogenetic tree indicates different group of clusters for each strain as depicted in (Fig. 1). The phylogenetic tree indicates different clusters for each isolate showing their sequence variation. The sequences obtained show greater than 96% similarity to the reference sequences available in NCBI GenBank database. Two reference sequences from genus *Aspergillus*, viz. *Aspergillus fumigatus* and *Aspergillus niger*, were used to draw outgroup to the existing clades generated by all study strains. Outgroup forms distinct clade distantly placed outside the clades formed by eleven strains.

Chrysosporium indicum (16.66%), the most dominant keratinophilic fungi in Indian subcontinent, was the most frequently isolated species. It is well adapted to warmer conditions of India [18]. *Microsporum gypseum* complex was the second most dominant fungi (11.53%). It has been reported from various parts of India [18–20]. *Chrysosporium tropicum* was

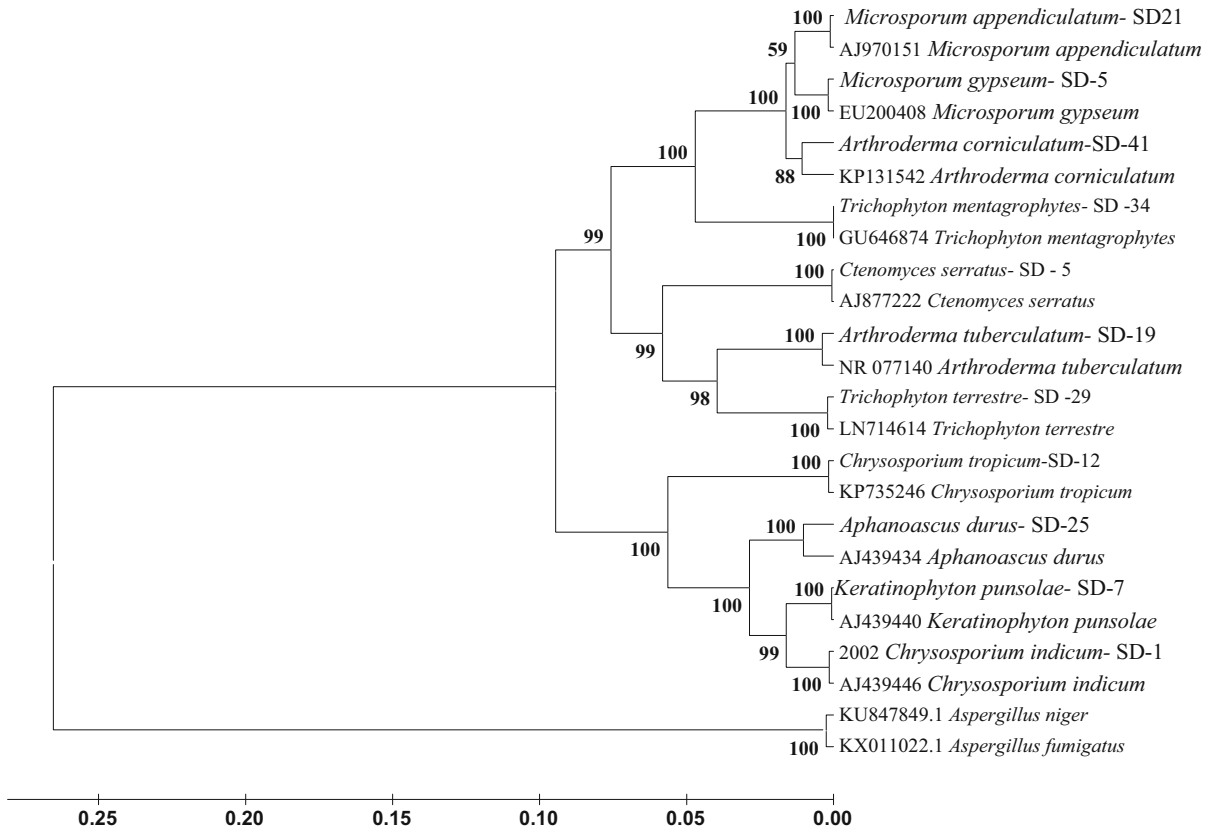

Fig. 1 Phylogenetic tree based on ITS1-5.8S-ITS4 region sequences for various isolated strains with reference strains. Numbers at the respective nodes are percentage of 1000 bootstrap replicates. *Bar* indicates genetic distance due to sequence variation. The tree was rooted *Aspergillus niger* and *Aspergillus fumigatus* as outgroup

3.84% in distribution. It is a cosmopolitan species and reported from various parts of India [21–23]. *Ctenomyces serratus* was recorded from 5.12% samples. It has been reported from various parts of India [24, 25].

The other fungi recorded were *Aphanoascus durus* (1.28%), *Keratinophyton punsolae* (1.28%), *Arthroderma tuberculatum* (3.84%), *Arthroderma corniculatum* (1.28%), *Microsporum appendiculatum* (1.28%), *Trichophyton mentagrophytes* (1.28%) and *Trichophyton terrestre* (2.56%).

Microsporum appendiculatum is reported for the first time from India. *Arthroderma corniculatum* was reported for the first time from the soils of Conakry as *Microsporon boulardii* [26]. Later on, its perfect state was reported as *Arthroderma corniculatum* [27]. It is also reported from Sanjay Gandhi National Park, Mumbai [7].

Trichophyton mentagrophytes is well-known dermatophyte and known to cause superficial infections to man and animals. *Trichophyton mentagrophytes* has been reported from soils at public places at Gulbarga [18], primary schools and public parks of Madras [28], public park of Mumbai [29]. Another species of *Trichophyton*, i.e., *T. terrestre*, was reported from soils of Agra [30], glacier bank soils [31] and salt pan soils of Mumbai [3].

Arthroderma tuberculatum was recovered from soils of caves around Mumbai [32], birds of Orissa [33] and also from pigeon feather from the buildings of Thane [34]. *Aphanoascus durus* was recorded from Indian soil of Gir Forest National Park and Wildlife Sanctuary, Gujarat [35], Sanjay Gandhi National Park, Mumbai [7] and soils of Lonar crater [24]. *Keratinophyton punsolae* was reported from the soil of Spain [13] and from the soil of Bahrain [4]. We are reporting for the first time the presence of keratinophilic fungi from Kaziranga National Park Assam (India).

Keratinophilic fungi play a vital role in nature in the breaking down and mineralization of keratinous

substrate into simpler substances. Their ability to grow on keratin regarded them as pathogens to human and livestock. For example, an invasive infection was noted in an 18-year-old woman who was a bone marrow transplant recipient as *Chrysosporium*, where the infection began as a facial swelling and extended into the central nervous system [36]. *Chrysosporium tropicum* was reported from comb lesion in two different breeds of chicken in India [37]. *Chrysosporium zonatum* was reported causing disseminated infection in a patient with chronic granulomatous disease [38]. In Japan, *C. zonatum* strains were also isolated from bronchial lavage from a female in Chiba and from a male in Kyushu. Both patients were with pulmonary cavities [39]. Recently, Chandrakar et al. [40] reported a case of subcutaneous fungal abscess over the great toe caused by a keratinophilic fungus, an unknown *Chrysosporium* sp., in a 60-year-old diabetic female. Thus, these fungi may be regarded as opportunistic pathogens.

Different *Chrysosporium* species have been identified in multiple reptile species that were presented with clinical signs of infection. *Chrysosporium guarroi* was described in pet green iguanas (*Iguana iguana*) from different geographic areas of Spain [41] and Turkey [42]. *Chrysosporium queenslandicum* has been reported in a garter snake [43]. *Chrysosporium keratinophilum*, *C. tropicum* and *Chrysosporium* spp. have been regarded as agents of cutaneous and systemic mycoses in reptile species, as reviewed by [44]. *Nannizziopsis vriesii* (CANV) has been identified as the cause of dermatitis in multiple reptile species such as chameleons [45], snakes [46, 47], salt-water crocodiles [48], bearded dragons [49, 50], green iguanas (*Iguana iguana*) [51] and girdled lizards [52]. *Nannizziopsis vriesii* was isolated from skin lesions in a pet bearded dragon [53], and *Chrysosporium ophiodiicola* was isolated from mycotic granuloma of a black rat snake [54]. *Aphanoascella galapagosensis* was recovered from carapace keratitis in a Galapagos tortoise residing in a south Texas zoological collection [55]. Most of the infections caused by these fungi have been described in the last 10 years and is unclear whether this could be attributed to recent climatic changes that could have affected the environment, where these animals live or that previous infections had been overlooked or misidentified [56]. Further studies and reports are needed to determine the importance of *Chrysosporium* spp. in captive and wild animals as well as the extent of its zoonotic capabilities.

References

1. Garg AP, Gandotra S, Mukerji KG, Pugh GJF. Ecology of keratinophilic fungi. Proc Ind Acad Sci (Plant Sci). 1985;94:149–63.
2. Vanbreuseghem R. Technique biologique pour 1' isolement des dermatophytes du sol. Ann Soc Belge Med Trop. 1952;32:173–8.
3. Deshmukh SK. Isolation of dermatophytes and other keratinophilic fungi from the vicinity of salt pan soils of Mumbai (India). Mycopathologia. 2004;157(3):265–7.
4. Deshmukh SK, Mandeel QA, Verekar SA. Isolation of keratinophilic fungi from the selected soils of Bahrain. Mycopathologia. 2008;165(3):143–7.
5. Javoreková S, Labuda R, Maková J, Novák J, Medo J, Majerčíková K. Keratinophilic fungi isolated from soils of long-term fold-grazed, degraded pastures in national parks of Slovakia. Mycopathologia. 2012;174(3):239–45.
6. Pakshir K, Ghiasi MR, Zomorodian K, Gharavi AR. Isolation and molecular identification of keratinophilic fungi from public parks soil in Shiraz, Iran. Biomed Res Int. 2013;2013:619576. doi:10.1155/2013/619576.
7. Deshmukh SK, Verekar SA. Isolation of keratinophilic fungi from selected soils of Sanjay Gandhi National Park Mumbai (India). Journal de Mycologie Médicale. 2014;24(4):319–27.
8. Anane S, Al-Yasiri MH, Normand AC, Ranque S. Distribution of keratinophilic fungi in soil across Tunisia: a descriptive study and review of the literature. Mycopathologia. 2015;180:61–8.
9. Sigler L, Carmichael JW. Taxonomy of *Malabranchea* and some other hyphomycetes with arthroconidia. Mycotaxon. 1976;4:349–488.
10. van Oorschot CAN. A revision of *Chrysosporium* and allied genera. Stud Mycol. 1980;20:1–89.
11. Currah RS. Taxonomy of onygenales: arthrodermaceae, gymnoascaceae, myxotrichaceae and onygenaceae. Mycotaxon. 1985;24:1–216.
12. von Arx JA. The ascomycetes genus *Gymnoascus*. Persoonia. 1986;13:173–83.
13. Cano J, Gurrao J. The genus *Aphanoascus*. Mycol Res. 1990;94:355–77.
14. Lee SB, Taylor JW. Isolation of DNA from fungal mycelium single cells. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ, editors. PCR protocols: a guide to methods and applications. San Diego, California: Academic Press; 1990. p. 282–7.
15. White TJ, Bruns T, Lee S, Taylor J. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis A, Gelfand DH, Sninsky JJ, White TJ, editors. PCR protocols: a guide to methods and applications. San Diego, California: Academic Press; 1990. p. 315–22.
16. Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ. Basic local alignment search tool. J Mol Biol. 1990;215:403–10.

17. Tamura K, Stecher G, Peterson D, Filipski A, Kumar S. MEGA6: molecular evolutionary genetics analysis version 6.0. Mol Biol Evol. 2013;30:2725–9.
18. Garg AK. Isolation of dermatophytes and other keratinophilic fungi from soils in India. Sabouraudia. 1966;4:259–64.
19. Singh CJ, Singh BG, Singh BS. Keratinophilic fungi of Ghana birds Sanctuary Bharatpur (Rajasthan). Ad Plant Sci. 1994;7:280–91.
20. Vidyasagar GM, Hosmani N, Shivkumar D. Keratinophilic fungi isolated from hospital dust and soils of public places at Gulbarga, India. Mycopathologia. 2005;159(1):13–21.
21. Carmichael JW. Chrysosporium and some other aleuriosporic hyphomycetes. Can J Bot. 1962;40:1137–73.
22. Deshmukh SK, Verekar SA. Incidence of keratinophilic fungi from the soils of Vedanthangal water bird sanctuary (India). Mycoses. 2011;54:487–90.
23. Randhawa HS, Sandhu RS. A survey of soil inhabiting dermatophytes and related kerationphilic fungi of India. Sabouraudia. 1965;4:71–9.
24. Deshmukh SK, Verekar SA. Keratinophilic fungi from the vicinity of meteorite crater soils of Lonar (India). Mycopathologia. 2006;162:303–6.
25. Deshmukh SK. Keratinophilic fungi isolated from soils of Mumbai, India. Mycopathologia. 1999;146:115–6.
26. Dominik T, Majchrowicz I. Some new species of fungi from the soil of Conakry. Mycopathol Mycol Appl. 1966;28:209–19.
27. Weitzman I, McGinnis MR, Padhye AA, Ajello L. The genus *Arthroderma* and its later synonym Nannizzia. Mycotaxon. 1986;25:505–18.
28. Ramesh VM, Hilda A. Incidence of keratinophilic fungi in the soil of primary schools and public parks of Madras City, India. Mycopathologia. 1998-99;143:139–145.
29. Deshmukh SK, Verekar SA. Prevalence of keratinophilic fungi in public park soils of Mumbai (India). Microbiol Res. 2012;3:e6. doi:10.4081/mr.2012.e6.
30. Saxena P, Kumar A, Shrivastava JN. Diversity of keratinophilic mycoflora in the soil of Agra (India). Folia Microbiol. 2004;49:430–4.
31. Deshmukh SK. Incidence of dermatophytes and other keratinophilic fungi in the glacier bank soils of Kashmir valley (India). Mycologist. 2002;16(4):165–7.
32. Verekar SA, Deshmukh SK. Incidence of keratinophilic fungi from caves around Mumbai, Maharashtra (India). J Basic Appl Mycology. 2006;5:62–4.
33. Sur B, Ghosh GR. Keratinophilic fungi from Orissa, India. II: Isolations from feathers of wild birds and domestic fowls. Sabouraudia. 1980;18:275–80.
34. Deshmukh SK. Keratinophilic fungi from feathers of pigeon in Maharashtra India. Mycoses. 2004;47:213–5.
35. Deshmukh SK, Verekar SA. Isolation of keratinophilic fungi from selected soils of The Gir forest national park and wildlife sanctuary, Gujarat, (India). Kavaka. 2014;43: 7–11.
36. Warwick A, Ferrieri P, Burke B, Blazar BR. Presumptive invasive *Chrysosporium* infection in bone marrow transplant recipient. Bone Marrow Transpl. 1991;8:319–22.
37. Saidi SA, Bhatt S, Richard JL, Sikdar A, Ghosh GR. *Chrysosporium tropicum* as a probable cause of mycoses of poultry in India. Mycopathologia. 1994;125:143–7.
38. Roilides E, Sigler L, Bibashi E, Katsifa H, Flaris N, Panteliadis C. Disseminated infection due to *Chrysosporium zonatum* in a patient with Chronic granulomatous disease and review of non-*Aspergillus* fungal infection in patients with this disease. J Clin Microbiol. 1999;37:18–25.
39. Sigler L, Flis AL, Carmichael JW. The genus *Uncinocarpus* (Onygenaceae) and its synonym *Brunneospora*: new concepts, combinations and connections to anamorphs in *Chrysosporium*, and further evidence of relationship with *Coccidioides immitis*. Can J Bot. 1998;76:1624–36.
40. Chandrakar S, Dias M, Shetty P, Tiewsoh JB, Guarro J. Isolation and characterization of an unknown *Chrysosporium* sp. producing subcutaneous mycosis in an immunocompromised patient. Mycopathologia. 2016;181:115–8.
41. Abarca ML, Castellá G, Martorell J, Cabañes FJ. *Chrysosporium guarroi* sp. nov. a new emerging pathogen of pet green iguanas (*Iguana iguana*). Med Mycol. 2010;48(2):365–72.
42. Kahraman BB, Sığırcı BD, Metiner K, Ak S, Koenhemsi L, Erman M, Castellá G, Abarca ML. Isolation of *Chrysosporium guarroi* in a Green Iguana (*Iguana iguana*), in Turkey. J Exot Pet Med. 2015;. doi:10.1053/j.jepm.2015.08.007.
43. Vissiennon T, Schuppel KF, Ullrich E, Kuijpers AF. Case report. A disseminated infection due to *Chrysosporium queenslandicum* in a garter snake (Thamnophis). Mycoses. 1999;42:107–10.
44. Paré JA, Jacobson E. Mycotic diseases of reptiles. In: Jacobson E, editor. Infectious diseases and pathology of reptiles: a color atlas and text. Boca Raton: CRC Press; 2007. p. 527–70.
45. Paré JA, Sigler L, Hunter DB, Summerbell RC, Smith DA, Machin KL. Cutaneous mycoses in chameleons caused by the *Chrysosporium* anamorph of *Nannizziopsis vriesii* (Apinis) Currah. J Zoo Wildl Med. 1997;28:443–53.
46. Bertelsen MF, Crawshaw GJ, Sigler L, Smith DA. Fatal cutaneous mycosis in tentacled snakes (*Erpeton tentaculatum*) caused by the *Chrysosporium* anamorph of *Nannizziopsis vriesii*. J Zoo Wildl Med. 2005;36:82–7.
47. Nichols DK, Weyant RS, Lamirande EW, Sigler L, Mason RT. Fatal mycotic dermatitis in captive brown tree snakes (*Boiga irregularis*). J Zoo Wildl Med. 1999;30:111–8.
48. Thomas AD, Sigler L, Peucker S, Norton JH, Nielan A. *Chrysosporium* anamorph of *Nannizziopsis vriesii* associated with fatal cutaneous mycoses in the salt-water crocodile (*Crocodylus porosus*). Med Mycol. 2002;40:143–51.
49. Bowman MR, Paré JA, Sigler L, Naeser JP, Sladky KK, Hanley CS, Helmer P, Phillips LA, Brower A, Porter R. Deep fungal dermatitis in three inland bearded dragons (*Pogona vitticeps*) caused by *Chrysosporium* anamorph of *Nannizziopsis vriesii*. Med Mycol. 2007;45:371–6.
50. Hedley J, Eatwell K, Hume L. Necrotising fungal dermatitis in a group of bearded dragons (*Pogona vitticeps*). Vet Rec. 2010;166:464–5.
51. Han J, Lee S, Na KJ. Necrotizing dermatomycosis caused by *Chrysosporium* spp. in three captive green ıguanas (*Iguana iguana*) in South Korea. J Exot Pet Med. 2010;19(3):240–4.
52. Hellebuyck T, Baert K, Pasmans F, Van Waeyenberghe L, Beernaert L, Chiers K, De Backer P, Haesebrouck F, Martel A. Cutaneous hyalohyphomycosis in a girdled lizard

(*Cordylus giganteus*) caused by the *Chrysosporium* anamorph of *Nannizziopsis vriesii* and successful treatment with voriconazole. Vet Dermatol. 2010;21:429–33.
53. Abarca ML, Martorell J, Castellá G, Ramis A, Cabañes FJ. Dermatomycosis in a pet inland bearded dragon (*Pogona vitticeps*) caused by a *Chrysosporium* species related to *Nannizziopsis vriesii*. Vet Dermatol. 2009;20:295–9.
54. Rajeev S, Sutton DA, Wickes BL, Miller DL, Giri D, Van Mete M, Thompson EH, Rinaldi MG, Romanelli AM, Cano JF, Guarro J. Isolation and characterization of a new fungal species, *Chrysosporium ophiodiicola*, from a mycotic granuloma of a black rat snake (*Elaphe obsoleta obsoleta*). J Clin Microbiol. 2009;47(4):1264–8.
55. Sutton DA, Marín Y, Thompson EH, Wickes BL, Fu J, García D, Swinford A, de Maar T, Guarro J. Isolation and characterization of a new fungal genus and species, *Aphanoascella galapagosensis*, from carapace keratitis of a Galapagos tortoise (*Chelonoidis nigra microphyes*). Med Mycol. 2013;51(2):113–20.
56. Stchigel AM, Sutton DA, Cano-Lira JF, Cabañes FJ, Abarca L, Tintelnot K, Wickes BL, García D, Guarro J. Phylogeny of chrysosporia infecting reptiles: proposal of the new family Nannizziopsiaceae and five new species. Persoonia. 2013;31:86–100.