

Case report

Oral Abscess Caused by *Chryseobacterium indologenes* in Ball Python (Python Regius); A Case Report

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Abstract: *Chryseobacterium indologenes* is an opportunistic pathogen isolated from human infections and rarely from some aquatic animals. A 3-year-old male ball python (*Python regius*) was admitted to the veterinary clinic by a pet owner because of acute respiratory and swallowing failure. During physical examinations, oral secretions and abscesses were observed on the mouth cavity and throat of the animal. After microbiological analysis including isolation, identification, and 16S rRNA sequencing; *Ch. indologenes* was detected as the main cause of the oral abscess in this case. Phylogenetic relatedness analysis showed a close relationship between this isolate and other strains isolated from human infections. Antimicrobial susceptibility testing revealed that the isolate was multi-drug resistant. However, it was very sensitive to minocycline, ceftazidime, and tetracycline. The patient was treated by antibiotic therapy and completely recovered after two weeks. To our best knowledge, this is the first incidence of *Ch. indologenes* in an oral abscess in a ball python. As result we would consider this organism as an opportunistic animal pathogen with zoonotic potentiality.

Keywords: *Chryseobacterium indologenes*; Oral abscess; Ball Python

1. Introduction

Chryseobacterium species are gram-negative bacteria and have sporadically and ubiquitously been found in the environment. These bacteria have often been detected in water, soil, waste, food sources, domestic animals, and aquatic environments. It was indicated that *Chryseobacterium* species are emerging multi-drug resistant opportunistic nosocomial pathogens and causes of different serious infections in neonates, pregnant women and immunocompromised patients exposed to medical device and environmental contaminants [1]. Recent research indicated *Chryseobacterium* as causes of some animal infectious diseases outbreaks in fish and aquatic animals. *Ch. balustinum*, *Ch. scophtalmum*, *Ch. joostei*, *Ch. piscicola* and *Ch. arothri* have been isolated from some fish species such as salmon and rainbow trout [2].

Ch. indologenes have also been reported as the main infectious agent of fatal diseases in newborns and other *Chryseobacterium* infections in human [3]. However, *Ch. indologenes* are rarely isolated in some aquatic species such as yellow perch, Mediterranean limpets, mussel, and purple sea urchins. The isolates were mainly recovered from liver, kidney, gills, and skin lesions. In these previous studies, all isolates were multi-resistant to a wide range of antibiotics [4, 5]. Investigation of genetic relatedness among the isolates showed the evolutionary patterns of the emerged pathogens [6]. Isolation of *Chryseobacterium* species such as *Ch. indologenes* from terrestrial sources has not been reported yet. At the present study, we investigated the first case of an infected ball python (*Pythonidae*) with an oral abscess, which, to our knowledge is the first case of *Chryseobacterium* infection in terrestrial animals, caused by multidrug resistant *Ch. indologenes*.

2. Case presentation

A three-year-old, 22 kg, male ball python (*Python regius*) was submitted by a pet owner to the Veterinary Clinic at Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran due to respiratory failure, difficulty swallowing, oral secretions, bad breath, anxiety and anorexia. The snake had been fed with commercial pet food and kept in a terrarium. The temperature and relative humidity in the terrarium were 35 °C and 80% RH. Clinical assessments of the snake were performed by the veterinarian of the center. Respiratory problem, bad breath, oral secretions, and several acute abscesses were observed in the mouth and throat of the snake by the veterinarian. Abscess samples were collected for microbiological analysis and isolation of the infectious agents of oral abscesses presented in the infected snake by oral surgery (Figure 1).



Figure 1. Oral abscess sample separated from the Ball Python mouth

Gram-negative bacilli were isolated from the abscess samples and the identities of the isolates were confirmed by biochemical tests. Antibiotic susceptibility of the isolates was evaluated using Kirby-Bauer disk diffusion described by the Clinical and Laboratory Standards Institute [7]. Biochemical analysis indicated that the isolate was beta-hemolytic; non-motile; indole positive; citrate negative; both methyl red and Voges-Proskauer's

negative; glucose was fermented with gas production; negative for urea, and unable to grow on MacConkey agar at 42 °C. Antibiotic susceptibility testing showed that the isolate was sensitive to minocycline, ceftazidime, and tetracycline; intermediate-sensitive to amikacin, ceftriaxone, ciprofloxacin, enrofloxacin, and levofloxacin; and resistant to imipenem, meropenem, penicillin, gentamycin, erythromycin, and azithromycin antibiotics. 16s rRNA gene sequencing was performed for definite species identification and construction of the phylogenetic tree of the isolate. 16s rRNA gene of the isolate was amplified by conventional polymerase chain reaction (PCR) using the universal primers pA (5'-AGA GTT TGA TCC TGG CTC AG-3') and pH (5'-AAG GAG GTG ATC CAG CCG CA-3')[8]. PCR products were extracted, purified, and sequenced by GATC company (Cologne, Germany). Sequencing of 16s rRNA gene (NCBI, GenBank accession number: MT276587) indicated that the isolate was *Chryseobacterium indologenes*. CLC software (CLC Genomic WorkBench, Qiagen, Hilden, Germany) was used to generate the cladogram by neighbor-joining clustering algorithm with bootstrapped 1000 times (using 16s rRNA gene sequence of the isolate and the NCBI GenBank database). Phylogenetic relationship among the *Ch. indologenes* isolated at the present study and the strains which were previously isolated in other studies is illustrated in Figure 2. Comparative 16s rRNA gene sequence analysis revealed high phylogenetic relatedness and sequence similarity between the 16s rRNA gene sequence of the strain, at the present study, isolated from oral abscess in python and those of the *Ch. indologenes* strains isolated from human infections in previous studies.

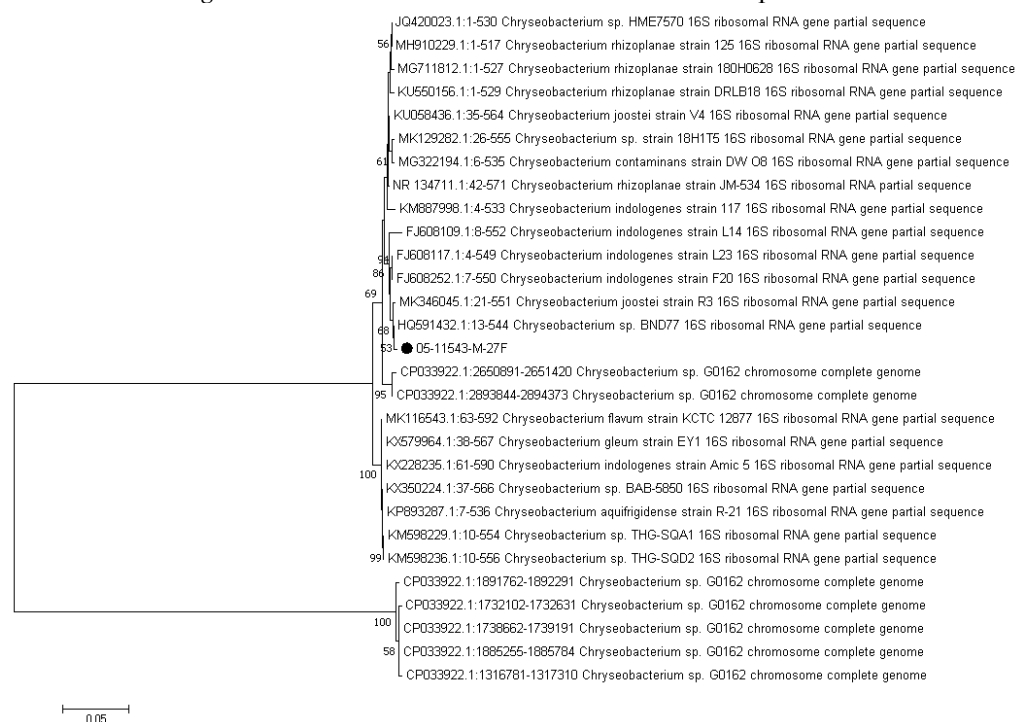


Figure 2. Phylogenetic tree based on 16s rRNA sequencing and neighbor joining method classification among the strain isolated in this report (NCBI, GenBank accession number: MT276587) and the strains isolated from human reported by other researchers published in NCBI

Ch. indologenes isolated from the oral abscesses in the python was sensitive to minocycline, ceftazidime, and tetracycline. The patient was treated with antibiotic therapy using tetracycline in a dose of 20 mg.kg⁻¹ every 48 hours for 14 days. During the treatment, the patient was kept in a terrarium and was fed by normal snake pet food. All equipment were disinfected with 10% formalin after each experimental session. After 2 weeks the patient was examined physically and microbiologically. Neither respiratory nor swallowing failure associated with the disease was observed in the patient. All oral and throat abscesses were treated and removed, leading to a complete recovery. Regarding the clinical results of the present study, minocycline, ceftazidime, and tetracycline antibiotics

have been recommended to be used as the main treatment of oral abscesses caused by *Ch. indologenes* in pythons.

3. Discussion

Ch. indologenes is an opportunistic and nosocomial pathogen and has been recorded threatening the health of neonates, immunocompromised patients, and patients with indwelling catheters. Most recently, *Ch. Indologenes* has been identified in a 52-year-old patient with end stage renal disease (ESRD) [9]. Multi-drug resistant nosocomial and opportunistic pathogens have been regarded as the major challenge among infectious diseases around the globe and have the potential to spread quickly [10]. *Ch. indologenes* have previously been isolated from some disease's aquatic animals such as salmon and rainbow trout fish [5]. There have not been any studies that reported *Chryseobacterium* species as the cause of oral or throat abscesses in human and animals as reported here. As part of the commercial animal trade, pythons are kept in zoos, animal parks, or as domestic pet and should be kept in a terrarium under appropriate temperature and relative humidity conditions [11]. In the case of the present study, the pet owner had kept the python in a suitable terrarium and fed with standard commercial pet food and water.

There are very limited reports and studies about the oral or mouth abscess in snakes. Lee and Kim (2011) reported Methicillin Resistant Staphylococcus aureus (MRSA) as the main cause of sub-spectacular abscess in a snake with respiratory failure symptoms. MRSA is a well-known zoonotic gram-positive pathogen around the world which has been reported as the major cause of serious abscess in many animal cases and a public health concern; consequently, detection and isolation of this pathogen from abscesses would be probable [12]. Another case study reported by Kurniawan and Govendan (2020) was about a Burmese Python with sub-spectacular abscesses. They found oral cavity inflammation and respiratory problems in the patient. However, no microbiological test was performed for the detection of abscess-causing pathogens and the infection was treated by antibiotics including oxytetracycline, meloxicam and enrofloxacin successfully [13]. *Ch. indologenes* have often been detected in infections with fatal outcomes in neonates. Also, there are several studies about isolation of this pathogen from immunocompromised patients, surface of the dwelling devices; and hospital vials, tubes, tap water, and environments as a nosocomial pathogen [14]. Close genetic relatedness between the isolate in this report and the isolates from humans reported in other case studies indicated a pathogenic potential of *Ch. indologenes* isolate in this study for humans.

Antimicrobial resistance patterns of the isolate are different from those of the isolates obtained from human infections reported by other researchers. The isolate found here was more sensitive than nosocomial isolates of *Ch. indologenes* (from human infections) to different antibiotics. Generally, non-nosocomial isolates showed fewer resistance to the antibiotics as we have observed in the present report. However, resistance to imipenem and meropenem antibiotics has been observed for both nosocomial and non-nosocomial *Ch. indologenes* isolates [15].

4. Conclusions

It is worthy to note that some pets may asymptotically transmit some pathogens to humans causing lethal emerging infectious diseases. These pets can be considered as an important source of emerging zoonotic diseases especially when a pathogen leads to infectious diseases such as abscesses or meningitis in both animals and humans [16]. However, in this report, we observed *Ch. indologenes* as the cause of an oral abscess in a pet python. Due to its differing antimicrobial susceptibility pattern, it may be possible that this isolate does not have the potential to be a human pathogen. It is suggested that for future studies, the antibiotic resistance and virulence factor encoded genes of *Ch. indologenes* isolates from both human and animal infections are comprehensively investigated to further understand the microorganism's role and an emerging zoonotic human pathogen.

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Conflicts of Interest: The authors declare that they have no competing interest.

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