Risk for Mycobacterium celatum Infection from Ferret

To the Editor: Mycobacterium celatum belongs to the group called “mycobacteria other than tuberculosis”; it is characterized by slow growth and a slender, rod-shaped form (0.25–0.5 × 0.5–13.0 μm). The cells are acid fast and do not form cords or branches. The species name, celatum, which means hidden or concealed, refers to the problem of phenotypically distinguishing the species from other mycobacteria, especially M. xenopi. M. celatum was first described in 1993 as a pathogen in persons with AIDS (1). Until now, few cases in humans have been reported; those cases were predominantly disseminated mycobacteriosis in immunocompromised patients (mainly those with AIDS), but they have also occurred in immunocompetent persons (1,2). For animals, 1 case of M. celatum infection in a ferret has been described (3). We describe another case in a ferret, with possible transmission to a human.

In 2009, a 3-year-old, neutered male, domestic ferret was examined in a veterinary clinic in Nuremberg, Germany, for a 5-month history of diarrhea. A chest radiograph showed multiple nodular densities in the lungs. Because of a poor prognosis, the ferret was euthanized. Necropsy was performed at the Institute of Veterinary Pathology in Munich. The lungs contained multifocal firm, light brown nodules, 6–10 mm in diameter (Figure, panel A). Spleen and lymph nodes (cervical, retropharyngeal, bronchial, gastric, mesenterial, popliteal) were enlarged. Histologic examination of lung, lymph nodes, spleen, liver, and brain showed granulomatous inflammation with predominantly macrophages, epithelioid cells (in the lung, including bronchioloes), and some multinucleated giant cells. Several acid-fast bacilli were visible with Ziehl-Neelsen staining, mainly intracytoplasmically in epithelioid cells (including those of bronchioloes) (Figure, panel B).

Conventional mycobacterial culture and PCR were used to look for mycobacteria in the lung, spleen, and lymph nodes. For culture, the material was homogenized, decontaminated, and spread onto solid Loewenstein-Jensen agar and injected into a liquid culture (Mycobacteria Growth Indicator Tube; Becton Dickinson, Heidelberg, Germany) for automated detection of mycobacterial growth.

DNA was extracted from the homogenized tissue by using the QiAmp DNA Mini Kit (QIAGEN, Hilden, Germany), and a 510-bp fragment at the 5′ end of the ribosomal 16S rDNA was amplified as described (4). The amplified fragment of the expected length was sequenced, and data were analyzed by using the Integrated Database Network System (SmartGene Services, Lausanne, Switzerland; www.smartgene.com). The resulting sequence was clearly interpretable and unambiguously assigned to M. celatum; sequence identity to GenBank accession no. Z46664 was complete except for 1 mismatch in bp 490. Minor sequence diversity in the M. celatum 16S rDNA gene has been documented (5). The most closely related species, M. kyorinense, differs substantially, having 11 mismatches within the 16S rDNA gene (6). Species identity was

References


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further supported by phylogenetic analyses of the hsp65 (7), rpoB (8), and sodA genes (9).

After 14 days of incubation, the solid and liquid cultures showed growth of acid-fast bacilli. Further identification at the Mycobacteria Reference Laboratory of the Bavarian Health and Food Safety Authority (Oberschleissheim, Germany) confirmed the molecular species typing results of M. celatum.

In Europe, naturally occurring mycobacterial infections in ferrets are rare; but in New Zealand, M. bovis or M. avium complex infections in ferrets are common (10). For ferrets, clinically relevant mycobacteria species are M. genavense and M. microti, among others. In humans, M. celatum mostly affects immunocompromised hosts with developing pneumonia or disseminated mycobacteriosis (1); inflammation in infected immuno-competent hosts is usually limited to the lungs or lymph nodes (2).

The ferret reported here had disseminated mycobacteriosis with no evidence of immunosuppression. In this respect, the clinical response of ferrets seems to differ from that in humans. The question of zoonotic risk remains. The ferret’s owner had had a cough for a long time, but radiography and sputum analysis showed no evidence of infection. However, these findings do not rule out mycobacteriosis because the owner had received unknown antimicrobial drug therapy before the samples for microbiology were collected. In general, potential transmission of mycobacteria should not be underestimated; in this case, intracytoplasmic bacilli were detected in the bronchioli of the ferret’s lung, making airborne spread of mycobacteria and infection of humans possible. The source of the primary infection in the ferret is not clear, but the dominant lesions in the lungs suggest it was airborne.

M. celatum infection must be strongly considered as a differential diagnosis in ferrets with pneumonia and generalized lymphatic hyperplasia. PCR and molecular species typing by 16S rDNA sequencing seem to be essential for an early and definitive diagnosis (1). The zoonotic risk for M. celatum infection in immunocompromised as well as immunocompetent persons should be kept in mind, considering the possible airborne transmission and the close contact between the animals and their owners.

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Human Infection with Pseudoterranova azarasi Roundworm

To the Editor: Eating raw or undercooked marine fish may lead to infection with several helminths. Members of the *Pseudoterranova decipiens* species complex are the second most common nematodes found in humans (most common are nematodes of the *Anisakis simplex* complex) (1,2). The *P. decipiens* species complex consists of at least 5 sibling species (genetically but not morphologically distinguishable): *P. decipiens* sensu stricto, *P. azarasi*, *P. cattani*, *P. krabbei*, and *P. bulbosa*. In northern Japan, human infection with *Pseudoterranova* spp. is not rare; by the mid-1990s, as many as 769 cases had been reported (3). Pseudoterranovaosis has also been encountered in North and South America and Europe (4,5). However, possible biologic and geographic differences of the sibling species of genus *Pseudoterranova* in relation to human infection remain unknown. We report a case of pseudoterranovaosis for which the sibling species was confirmed as *P. azarasi*.

In 2009, a woman in Japan coughed up a nematode and expelled it through her mouth. Her medical history was unremarkable, and she had not traveled abroad for the past few years. Measurements of the worm were as follows: body 35 mm long and 0.85 mm wide; esophagus 1.88 mm long; ventriculus 1.05 mm long; and intestinal cecum 0.95 mm long, extending anteriorly along the ventriculus. The anterior end of the worm contained 3 lips. The tail was conical, 0.21 mm long, and had a small, knob-like process at the posterior end. On the basis of morphologic features, the worm was identified as a 4th-stage larva of *P. decipiens* (sensu lato) roundworms.

DNA was extracted from the isolate from the patient (clinical isolate) and from isolates of *P. decipiens* (sensu lato) larvae from Pacific cod purchased at a local market. PCR was performed and the amplification products were directly sequenced. Primers used were 5′-CCGCGACAAA AGTCGTAACAA-3′ and 5′-ATATG CTTAAATTGCAGGT-3′ for a region that spans the internal transcribed spacer (ITS) 1, ITS2, and 5.8S rRNA; 5′-CTACTAAGAATTGTGC-3′ and 5′-AATCCAAATCCTACGG AGA-3′ for cytochrome oxidase subunit 1; and 5′-CAGCCTATGGCTCT ACTAA-3′ and 5′-AGCATAACAA AGAACTCA-3′ for NADH dehydrogenase subunit 1.

Nucleotide sequences for the clinical and Pacific cod isolates (GenBank accession nos. AB576756–AB576761) were compared with those in DNA databases available to the public. The ITS1 and ITS2 sequences of the clinical and Pacific cod isolates were identical to those of *P. azarasi* roundworms; however, the ITS sequences of these 2 isolates also showed close similarity to those of *P. decipiens* (sensu stricto) worms and differed by only 1 nt in ITS2. However, phylogenetic tree analyses of NADH dehydrogenase subunit 1 and cytochrome oxidase subunit 1 sequences showed that the clinical and Pacific cod roundworm isolates clustered with *P. azarasi* and were clearly distinguished from *P. decipiens* (sensu lato) roundworms.

Geographic distribution of the 5 sibling species of the *P. decipiens* complex differs somewhat. *P. azarasi* and *P. bulbosa* are found in northwestern Pacific (including Japan), *P. decipiens* (sensu stricto) and *P. krabbei* in northeastern Atlantic, *P. decipiens* (sensu stricto) in northwestern Atlantic, and *P. cattani* in southeastern Pacific waters (6). Given this distribution, it is not surprising...