Niacin-Positive \textit{Mycobacterium kansasii} Isolated from Immunocompromised Patients

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Niacin-positive \textit{Mycobacterium kansasii} was isolated from three patients, two with respiratory infections and one with a perirectal abscess. The isolates were phenotypically similar to other strains of \textit{M. kansasii}, differing only in their ability to produce niacin. This phenotype has been reported only twice in the literature, during the 1960s.

\textit{Mycobacterium kansasii} has been reported previously to cause both pulmonary and extrapulmonary infections \cite{2, 14, 15}. In addition to chronic pulmonary disease, \textit{M. kansasii} may cause cervical lymphadenitis, dermatitis, osteomyelitis, and arthritis. Disseminated infection occurs most commonly in immunocompromised patients \cite{10}. The organism can be identified in the laboratory on the basis of pigment and biochemical studies: \textit{M. kansasii} is usually photochromogenic (rare isolates may be nonpigmented or scotochromogenic), and niacin accumulation is thought to be uniformly negative for this species. However, we recently isolated niacin-producing isolates of \textit{M. kansasii} from three of our patients with compromised immune function, one at the University of Pennsylvania and two at the University of Cincinnati (two patients had respiratory infections, and one patient had a perirectal abscess).

\textbf{Case 1 (University of Pennsylvania).} A 34-year-old black man, known to be positive for human immunodeficiency virus (HIV) antibody since August 1988, presented in September 1989 with a cough productive of blood-streaked sputum, increasing shortness of breath, pleuritic chest pain, and low-grade fevers. Diffuse interstitial bilateral infiltrates were observed on the chest radiograph. Bronchial washings showed many acid-fast bacilli by fluorochrome and Kinyoun staining and \textit{Pneumocystis carinii} on silver staining. He was treated with isoniazid (INH), rifampin, and ethambutol for mycobacterial infection and cotrimoxazole and methylprednisolone for pneumocystis infection, with clinical improvement. A follow-up chest radiograph showed cavitation of the left upper lung infiltrate, with resolving interstitial infiltrates. He improved clinically and was discharged after 3 weeks of treatment for \textit{P. carinii} pneumonia with zidovudine, INH, rifampin, and ethambutol. The isolated \textit{Mycobacterium} sp. was subsequently identified as niacin-positive \textit{M. kansasii}. After 1 year of treatment for \textit{M. kansasii} pneumonia, his left apical lung cavity resolved and his antituberculous therapy was discontinued.

\textbf{Case 2 (University of Cincinnati).} A 37-year-old white man who had been HIV positive for several years developed a cough, followed by shortness of breath and left anterior chest pain, in May 1990. A chest radiograph showed bilateral lower lobe minimal pneumonia. Bronchoscopy specimens were negative for \textit{P. carinii}, fungi, and bacteria. However, multiple acid-fast organisms were identified from a transbronchial biopsy from the superior segment of the right upper lobe, obtained because of a nodular intrabronchial lesion seen on bronchoscopy. Cultures of the biopsy grew a \textit{Mycobacterium} sp. subsequently identified as niacin-positive \textit{M. kansasii}. Initially, he was treated with clofazimine, ethambutol, rifampin, and ciprofloxacin for presumed \textit{Mycobacterium avium} complex infection. After identification of the mycobacterial isolate, his therapy was changed to INH, ethambutol, and rifampin. At present, the patient is doing well clinically and remains on therapy.

\textbf{Case 3 (University of Cincinnati).} A 51-year-old black man received his second cadaveric kidney transplant in August 1978. In July 1989, he developed significant weight loss, low-grade fever, night sweats, and pain around the rectum. The patient was negative for HIV antibody. A CT scan showed a small perirectal fluid collection, but no action was taken because symptoms improved. In November 1989, he experienced additional weight loss and generalized malaise. A repeat CT scan revealed a massive pelvic fluid collection and the appearance of a large perirectal abscess. Pus drained surgically from his perirectal abscess showed numerous acid-fast bacilli, which grew a \textit{Mycobacterium} sp. identified as niacin-positive \textit{M. kansasii}. He was treated with INH, rifampin, ethambutol, and prednisone after surgery. The surgical wound healed in an expected fashion and eventually closed without incident. Antimycobacterial therapy and maintenance immunosuppressive therapy have continued to the present, and the patient remains well.

Specimens at both institutions were processed by using the \textit{N}-acetyl-l-cysteine-\textit{NaOH} technique \cite{11, 13} and concentrated by centrifugation at 3,000 to 3,800 \( \times \) g. Specimens were inoculated to Lowenstein-Jensen (LJ) medium slants, Middlebrook 7H11 agar, and Mitchison 7H11 Selective Agar at the University of Pennsylvania and to the surface of tubed slants consisting of LJ medium and the Gruft modification of LJ at the University of Cincinnati. Media were incubated at 35°C in a 5 to 10% \textit{CO}_2 atmosphere. Biochemical tests were performed by using conventional methods \cite{13}. Niacin test-

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ing was performed by the paper strip method (Difco, Detroit, Mich.). All specimens examined from these patients showed the presence of acid-fast bacilli by fluorochrome staining. A number of biochemical and growth studies were performed on the isolates (Table 1). Growth on primary media was noted after 2 1/2 to 3 weeks of incubation, and colonies exhibited photochromogenicity. Tests for niacin accumulation on all isolates were positive. Several biochemical tests, including nitrate reduction and semiquantitative catalase tests, were useful in distinguishing our isolates from Mycobacterium marinum, and in addition, Tween hydrolysis and arylsulfatase tests were helpful in distinguishing the isolates from Mycobacterium simiae. Growth temperature studies are also useful for distinguishing M. kansasii from M. marinum. The isolates were tested for susceptibility to INH, ethambutol, streptomycin, and rifampin by the proportion method of susceptibility (13). All three isolates were partially INH resistant at 0.2 μg/ml, ethambutol susceptible (5.0 μg/ml), and rifampin susceptible (1.0 μg/ml). All isolates were susceptible to streptomycin at 10 μg/ml, but only two of the three were susceptible to streptomycin at 2.0 μg/ml.

Additional confirmatory tests were performed. M. kansasii produces a unique N-acylaminogluco sugar in the lipooligosaccharide, named N-acylmanosamine (7), and is unique among the different mycobacterial species. By using thin-layer chromatography, the lipooligosaccharides isolated from our three isolates were shown to be alkali labile, and a thin-layer chromatography pattern consistent with M. kansasii, and, by using a specific monoclonal antibody directed against N-acylmanosamine, also reacted in an enzyme-linked immunosorbent assay (ELISA) (4, 8). Intact lipid was used in an ELISA using a monoclonal antibody directed against N-acylmanosamine (8) with ELISA optical density values ranging from 0.986 to >2.0. Lipid derived from a negative control strain, M. avium complex, showed a negative reaction.

Niacin-positive M. kansasii have been reported only twice in the literature, during the 1960s. Yue and Cohen (16) reported one fatal case of pulmonary infection due to niacin-positive M. kansasii in a 71-year-old man with chronic rheumatoid arthritis who developed bilateral cavitary lung disease. Acid-fast staining of lung tissue was negative, but cultures grew the organism. Premortem sputum cultures also grew the organism on several occasions. It was streptomycin resistant and partially resistant to INH and PAS. Gimpel et al. (6) also reported the isolation of a niacin-positive strain of M. kansasii from a laryngeal swab of a patient with pulmonary disease. The organism was resistant to INH, streptomycin, and PAS. Clinical findings about this patient were not reported.

Although niacin accumulation is commonly used for the identification of Mycobacterium tuberculosis, other mycobacteria can produce niacin. M. marinum and M. simiae may give a positive reaction but can be differentiated from M. kansasii on the basis of growth temperature studies and selected biochemical tests (11). M. kansasii also contains an alkali-labile lipooligosaccharide (3). A unique, N-acylaminogluco sugar epitope is present in M. kansasii which was described by Hunter et al. (7, 8) and detected in our isolates by ELISA. M. tuberculosis, Mycobacterium bovis, M. simiae, and M. marinum do not contain this epitope.

Infection with M. kansasii is common in immunocompromised patients and is thought to be the second most common nontuberculous mycobacterial infection in patients with AIDS (9). Of interest is a recent report of 19 AIDS patients with M. kansasii infection in which it was found that 6 patients had concurrent P. carinii infections, similar to the situation of one of our patients. The finding of cavi
tary disease in case 1 is consistent with its role as a significant pathogen and is a good predictor of M. kansasii disease in patients without AIDS (1). M. kansasii has been reported to cause abscesses in patients with underlying immunodeficiency, such as in HIV infection (12) and renal transplantation (5). Although M. kansasii has been isolated from different body sites, case 3 is the first reported instance, to our knowledge, of M. kansasii causing a perirectal ab-

The susceptibility patterns of the three isolates were consistent with those of the usually nai-
cin-negative M. kansasii strains. This finding and the fact that antimicrobial therapy was effective in our patients suggest that niacin positivity was not associated with any change in the viru-

Table 1. Biochemical tests useful in differentiating M. kansasii from other mycobacteria that may exhibit niacin accumulation

<table>
<thead>
<tr>
<th>Species</th>
<th>Niacin</th>
<th>Nitrate reduction</th>
<th>Semiquantitative catalase</th>
<th>68°C catalase</th>
<th>Arylsulfatase (14 days)</th>
<th>Tween hydrolysis</th>
<th>Urease</th>
</tr>
</thead>
<tbody>
<tr>
<td>M. kansasii*a</td>
<td>+</td>
<td>+</td>
<td>+</td>
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<tr>
<td>M. kansasii*b</td>
<td>+</td>
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<td>+</td>
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<tr>
<td>M. simiae*c</td>
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*a Data are from the present study.
*b Data are from reference 11.
ADDENDUM IN PROOF

Subsequent to the above studies, additional isolates with the niacin-positive phenotype have been isolated at the University of Pennsylvania (n = 1) and the University of Cincinnati (n = 2).

REFERENCES