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ORIGINAL INVESTIGATION

Clinical and Epidemiologic Features of Infection With *Mycobacterium genavense*

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Objectives: To characterize clinical and epidemiologic features of infections with *Mycobacterium genavense*.

Design: Case series and case-control studies. Patients with *M genavense* were compared with two control groups: CD4 controls were matched on the basis of CD4 counts, and *Mycobacterium avium-intracellulare* complex controls had disseminated infection with *M avium-intracellulare* complex.

Results: Fifty-four patients with disseminated infections caused by *M genavense* were found, from Europe (37), North America (15), and Australia (two). All were infected with human immunodeficiency virus. The median CD4 count was 0.016×10^9/L (16/mm^3) (range, 0.001 to 0.082×10^9/L). Eighty-seven percent had fever and weight loss, 44% had diarrhea, 43% had splenomegaly, 39% had hepatomegaly, and 72% had anemia. In Swiss university hospitals, *M genavense* was responsible for 12.8% of nontuberculous disseminated mycobacterial infections in patients with human immunodeficiency virus from 1990 to 1992. The median survival was 190 days after the first isolation of *M genavense*. Among the patients who had been treated with at least two antimycobacterial drugs for 1 month or more, median survival was 263 days (95% confidence interval, 144 to 382 days), compared with 81 days (95% confidence interval, 73 to 89 days) for those not treated (P=.0009). Survival in patients with *M genavense* was similar to the survival of *M avium-intracellulare* complex controls. However, patients with similar CD4 counts (CD4 controls) survived longer (median, 342 days; 95% confidence interval, 269 to 415 days; P<.0003).

Conclusions: Infection with *M genavense* may be responsible for more than 10% of disseminated nontuberculous mycobacterial infections in patients with human immunodeficiency virus infection. Its clinical presentation and response to treatment are similar to those of infection with *M avium-intracellulare* complex.

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MYCOBACTERIAL infections are a frequent complication of human immunodeficiency virus (HIV) infection and impair both survival and quality of life.[1] After a diagnosis of acquired immunodeficiency syndrome (AIDS), the incidence of *Mycobacterium avium-intracellulare* complex (MAC) bacteremia was 21% at 1 year and 43% at 2 years.[2] In a retrospective study of 1984 cases of disseminated mycobacteriosis, invasive MAC infection was found in 1906 (96%), while *Mycobacterium kansasii, Mycobacterium gordonae, Mycobacterium fortuitum,* and *Mycobacterium chelonae* accounted for the rest.[3] However, these data may be biased in favor of mycobacterial infections that are easy to diagnose and mycobacteria that are easily cultured.

Mycobacterium genavense is a newly recognized fastidious pathogen that does not grow on conventional solid media,[4,5] although limited growth has been observed in liquid (Middlebrook 13) culture. The first patient suffered from fever and gastrointestinal tract symptoms.[6] Acid-fast rods were found in the duodenum, feces, urine, and bone marrow and in multiple tissues at autopsy. Electron microscopy and lipid analysis suggested that the pathogenic agent was a mycobacterium. The infection could be transferred to athymic nude mice but not to immunocompetent mice. Amplification and sequencing of the 16S ribosomal RNA gene demonstrated that the pathogen was a new species, which was later named *M genavense*.[7,8] In this study, we describe the clinical and epidemiologic features in 54 patients with AIDS with disseminated infection caused by *M genavense*, and we use case-control methods to compare patients with *M genavense* with other patients with HIV infection.

See Patients and Methods on next page

PATIENTS AND METHODS

For a patient to be included in this series, at least one culture of blood (n=50), bone marrow (n=2), or biopsy tissues (n=2) had to be positive for *M genavense*. Some of these cases have been partly described previously.[8-14] Patients with only pulmonary or gastrointestinal tract colonization were excluded. Clinical information came from a standard questionnaire. The date of occurrence of *M genavense* or MAC was defined as the time when the first positive sample was taken. The definition of AIDS was that of the criteria established in 1987 by the Centers for Disease Control and Prevention.[15] The diagnosis of *M genavense* infection was established by polymerase chain reaction amplification and sequencing of the 16S rRNA gene as previously described.[8,16-18]

Cases of infection with *M genavense* were compared with two control groups. The first included patients with disseminated MAC infections (MAC controls). For each case infected with *M genavense*, one control patient was chosen from the same facility, who had blood cultures positive for MAC within 6 months before or after *M genavense* was found in the case patient. The second control group included patients with a similar degree of immunosuppression as measured by the CD4 count, but without documented *M genavense* infection (CD4 controls). For each case with *M genavense*, control patients were chosen who were matched for hospital or region of origin and whose CD4 counts were comparable with that of the case patient (the difference between the counts in the case patient and controls varied from 0 to 0.050×10^9/L [0 to 50/mm^3]) within 1 month of diagnosis of *M genavense* infection. Controls either had blood cultures that were negative or contained bacteria other than *M genavense*, or did not have blood cultures. For each case, two controls were chosen (with the exception of two cases for which only one control was available). A standard questionnaire was filled out for all MAC controls and CD4 controls.

To compare the frequency of infections with *M genavense* with the frequency of disseminated mycobacterial infections in patients with AIDS, data from the Swiss HIV Cohort Study were used. This study contains data on more than 6000 HIV-positive patients treated at the Swiss university hospitals, with regular standardized laboratory and clinical examinations.[19]

Survival of cases and controls was analyzed by the Kaplan-Meier life table method and compared by the Mantel-Cox (log-rank) tests.[20]

RESULTS

Fifty-four cases of disseminated *M genavense* infection were identified from three continents. There were no known contacts between cases. The 37 European cases (68%) came from Switzerland (29), Germany (four), Italy (two), and Austria (two). Fifteen cases (28%) were diagnosed in North America in Seattle, Wash (eight); Denver, Colo (five); and Iowa City, Iowa (two). Two cases (4%) were from Melbourne, Australia. All patients were infected with HIV, and all risk groups were represented. There were 27 homosexual or bisexual men (50%), 15 injecting drug users (28%), two (4%) who had both risk factors, six (11%) who had acquired HIV by heterosexual transmission, three (6%) who had acquired HIV through blood products, and one (2%) who had acquired HIV through congenital transmission. In North America, all cases were male, and 13 (87%) of 15 were homosexual. The median age at the time of *M genavense* diagnosis was 34 years (range, 6 to 51 years). All patients were severely immunosuppressed, with a median CD4 count of 0.016×10^9/L (range, 0.001 to 0.082×10^9/L). Forty-six cases (85%) had CD4 counts of less than 0.050×10^9/L. In 16 patients (30%), *M genavense* was the first major opportunistic infection. In the remaining 38 patients (70%), AIDS had been diagnosed an average of 15.2 months earlier (range, 1.3 to 48.9 months). At the time of diagnosis, all patients were symptomatic (**Table 1**). The most common clinical signs were fever in 47 cases and weight loss in 47 cases (87%). Gastrointestinal tract symptoms and signs were frequent. Chronic diarrhea was found in 24 (44%), splenomegaly in 23 (43%), hepatomegaly in 21 (39%), and abdominal pain in 16 (30%). Hematologic abnormalities were also common, with anemia in 39 cases (72%) and pancytopenia in 17 cases (32%). Pulmonary symptoms were present in only six cases (11%) and were often associated with another pulmonary disease, such as *Pneumocystis carinii* pneumonia.

Table 1. Symptoms and Signs in Disseminated Infection With *Mycobacterium genavense* (N=54)

Symptoms/Signs	No. (%)
General (systemic)	
Fever	47 (87)
Weight loss	47 (87)
Gastrointestinal tract	
Pain	16 (30)
Diarrhea	24 (44)
Splenomegaly	23 (43)
Hepatomegaly	21 (39)
Blood	
Anemia	39 (72)
Pancytopenia	17 (32)
Others	
Pulmonary	6 (11)
Asymptomatic	0 (0)

To estimate the frequency of infection with *M genavense*, we counted cases with disseminated mycobacterial infections in general, and with *M genavense* in particular, diagnosed in 1990, 1991, and 1992 in the Swiss HIV Cohort Study. One hundred eighty-seven disseminated mycobacterial infections were recorded, of which 24 (12.8%) were caused by *M genavense*.

Two cases diagnosed at autopsy were excluded

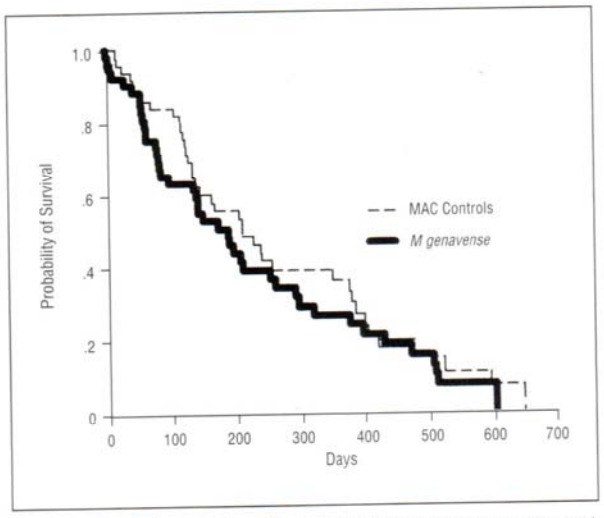

Figure 1. *Survival curves for patients with* Mycobacterium genavense *and* Mycobacterium avium-intracellulare *complex (MAC) controls.*

Table 2. Case-Control Study*

	Patients With *Mycobacterium genavense* (N=52)	MAC Controls (N=52)	CD4 Controls (N=98)
Sex, No.			
F	7	11	16
M	45	41	82
Risk group			
Homosexual	28	31	64
Heterosexual	6	8	11
Injecting drug abuse	14	11	18
Blood	3	1	1
Other	1	1	4
Age, y (average/median)	33.5/33.4	35.8/34.4	36.2/34.5
CD4×10^9/L (average/median)	0.025/0.016	0.022/0.010	0.025/0.016
Treatment of HIV, No.	36	29	70
Prophylaxis for PCP, No.	40	40	77

**MAC indicates* Mycobacterium avium-intracellulare *complex; HIV, human immunodeficiency virus; and PCP,* Pneumocystis carinii *pneumonia. None of the differences between the two groups were significant (two-tailed χ^2 test).*

from the survival analysis illustrated in **Figure 1**. Day 0 was defined as the day of the first sample that ultimately yielded *M genavense*, ie, usually the day the first positive blood culture was taken. The estimated mortality was 35% at 3 months, 51% at 6 months, and 76% at 12 months. Most patients had general and gastrointestinal tract symptoms at the time of death. According to the physicians in charge, *M genavense* infection was the principal or major contributing cause of death in 25 (56%) of the 45 cases who died. Two deaths were definitively not HIV related (suicide in one and extensive burns in one).

With a case-control study, we compared the prognosis of infections with *M genavense* and MAC. The MAC controls were matched for the date of diagnosis and the region of origin. **Table 2** shows the characteristics of the two groups. There were no significant differences in risk group, sex, age, or CD4 count. In both cases and MAC controls, the most frequent clinical presentations were systemic signs, hepatosplenomegaly, diarrhea, abdominal pain, and anemia. The proportion of patients who received antimycobacterial treatment (for more than 1 month with at least two drugs) was 54% for *M genavense* and 56% for MAC. Anti-HIV treatment and prophylaxis for *P carinii* pneumonia were given to 69% and 77% of the patients with *M genavense* and 73% and 77% of the patients with MAC. The median survival of patients with *M genavense* was 190 days (95% confidence interval [CI], 131 to 249 days) and that of MAC controls was 212 days (95% CI, 124 to 300 days), a nonsignificant difference (P=.37) (Figure 1).

Does infection with *M genavense* adversely affect the prognosis? We tried to answer this question by comparing patients with *M genavense* with patients with a comparable degree of immunosuppression as indicated by CD4 count. Two *M genavense* cases were excluded from this analysis because CD4 counts were not available. Characteristics of controls, matched for CD4 count, date when CD4 counts were determined, and region of origin are listed in Table 2. There were no significant differences between the two groups in risk behavior, sex, or age. Twelve CD4 controls were infected with MAC. The median survival was 190 days (95% CI, 123 to 257 days) for *M genavense* and 342 days (95% CI, 269 to 415 days) for CD4 controls (**Figure 2**). This was a significant difference according to the log-rank test (P<.0003).

At the time these patients were seen, there was no standard therapy for treating infection with atypical mycobacteria. Twelve patients were not treated, 41 patients received at least one antimycobacterial drug for at least 1 day, and 30 patients received at least two such drugs for more than 1 month. Drugs used are listed in **Table 3**. Survival analysis showed a difference between treated and untreated patients, with a median survival of 210 days for the group with any treatment and 81 days for the untreated group (P=.02, Cox-Mantel log-rank test). The difference was more striking if "treatment" was defined as receiving at least two antimycobacterial drugs for at least 1 month (**Figure 3**), with a median survival of 263 days (95% CI, 144 to 382 days) compared with 81 days (95% CI, 73 to 89 days) for the totally untreated patients who survived at least 1 month (log-rank test, P=.0009). Between treated and untreated patients, there were no significant differences in sex, age, risk groups, occurrence of other opportunistic infections, and CD4 count.

COMMENT

In Switzerland, *M genavense* was responsible for 12.8% of disseminated mycobacterial infections in patients with AIDS from 1990 to 1992. Among 30 consecutive instances where mycobacteria grew in liquid blood cultures, but not in standard subcultures, we diag-

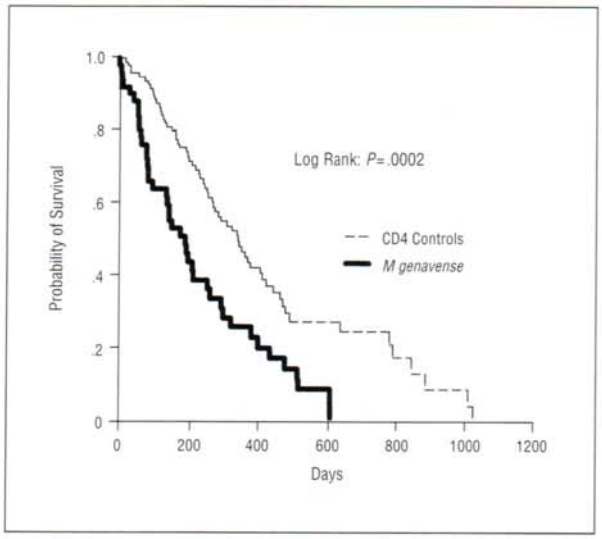

Figure 2. *Survival curves for patients with* Mycobacterium genavense *and CD4 controls.*

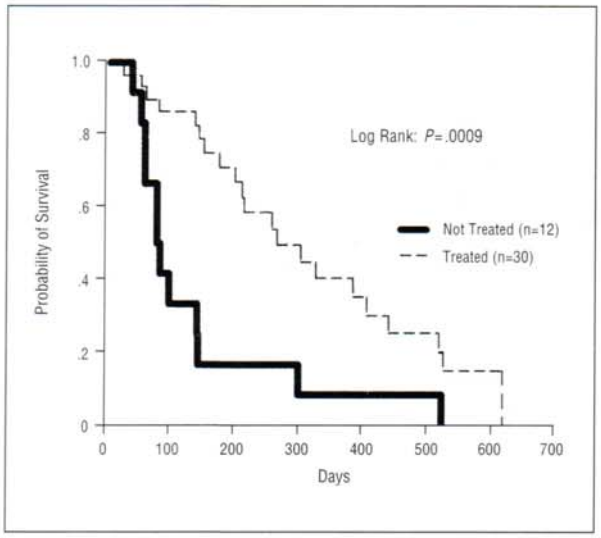

Figure 3. *Survival for treated and untreated patients with disseminated* Mycobacterium genavense *infection. Treatment was defined as having received at least two antimycobacterial drugs for at least 1 month. Patients who survived less than 1 month were excluded from analysis.*

Table 3. Drugs Used for Treating Patients With *Mycobacterium genavense* (n=41)

Drug	No. of Treated Cases (%)
Ethambutol	32 (78)
Rifampin	29 (71)
Clofazimine	23 (56)
Ciprofloxacin	21 (51)
Isoniazid	19 (46)
Clarithromycin	17 (41)
Amikacin	11 (27)
Pyrazinamide	9 (22)
Rifabutin	7 (17)

nosed 29 cases of *M genavense* infection and one case of MAC infection by polymerase chain reaction. In fact, it was found wherever practitioners who treat many patients with AIDS looked for it, most notably in Switzerland, but also elsewhere in Europe, in the United States, in Canada, and in Australia. Therefore, we assume that this microorganism is widespread. So far it has been isolated from humans and pet birds.[21] Although, in this series, all 54 patients had HIV infection, *M genavense* was also recently identified in an HIV-negative child with abdominal pain and enlarged abdominal lymph nodes (D. Turck, MD, Lille, France, personal communication, November 1993). All of our patients were severely immunosuppressed, with a median CD4 count of only 0.016×10^9/L. At the time of infection with *M genavense,* they were also exposed to many other opportunistic pathogens, MAC among them. Nonetheless, we did not find double infections with MAC and *M genavense.* Possibly, this is because of the higher growth rate of MAC in culture, which might obscure concomitant infection with *M genavense.* In one patient, MAC was found 2 months after *M genavense.*

Pathogenic mechanisms are still unknown. The first step leading to infection may be colonization of the gastrointestinal tract. Two patients who were not included in this series had *M genavense* in feces but negative blood cultures. Furthermore, gastrointestinal tract symptoms were frequent, and intestinal biopsy specimens, obtained clinically or at autopsy, showed massive infiltration of submucosal connective tissue. In contrast, we did not find evidence of pulmonary colonization or disease.

At the time of diagnosis, patients had advanced HIV infection with systemic, gastrointestinal tract, or hematologic signs or symptoms. We cannot prove that all these were caused only by *M genavense,* because many of the signs and symptoms are common in AIDS and may result from a number of other opportunistic diseases. Indeed, one may doubt whether infection with *M genavense* affects the prognosis. To answer this question, 50 cases were compared with control patients with a similar degree of immunosuppression as indicated by CD4 lymphocyte count. The survival was clearly diminished in the group with *M genavense* (Figure 2). Our interpretation is that *M genavense* lessens survival and that it is probably important to treat this infection. Comparison of patients with *M genavense* and matched patients with MAC disclosed comparable survival (Figure 1). Patients with MAC who are treated survive longer than those who are not.[22] The same appears to be true for *M genavense,* as we found a median survival of 263 days (95% CI, 144 to 382 days) in those who received at least a month of treatment compared with 81 days (95% CI, 73 to 89 days) in those who did not (Figure 3). It should be stressed that ours was a retrospective analysis, and the assignment to treatment was not randomized. Therefore, a bias toward treatment in patients

who are "healthier" cannot be excluded. Nevertheless, for the time being, we recommend treating *M genavense* infection in a manner similar to MAC. In the future, clinical trials, results of treatment in the nude mouse model, and characterization of resistance factors will indicate the best treatment strategy for patients infected with *M genavense*.

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REFERENCES

1. Horsburgh CR Jr. *Mycobacterium avium* complex infection in the acquired immunodeficiency syndrome. *N Engl J Med.* 1991;324:1332-1338.
2. Nightingale SD, Byrd LT, Southern PM, Jockusch JD, Cal SX, Wynne BA. Incidence of *Mycobacterium avium-intracellulare* complex bacteremia in human immunodeficiency virus–positive patients. *J Infect Dis.* 1992;165: 1082-1085.
3. Horsburgh CR Jr, Selik RM. The epidemiology of disseminated nontuberculous mycobacterial infection in the acquired immunodeficiency syndrome (AIDS). *Am Rev Respir Dis.* 1989;139:4-7.
4. Böttger EC, Hirschel B, Coyle MB. *Mycobacterium genavense* sp. nov. *Int J Syst Bacteriol.* 1993;43:841-843.
5. Coyle MB, Carlson LC, Wallis CK, et al. Laboratory aspects of '*Mycobacterium genavense*,' a proposed species isolated from AIDS patients. *J Clin Microbiol.* 1992;30:3206-3212.
6. Hirschel B, Chang HR, Mach N, et al. Fatal infection with a novel unidentified mycobacterium in a man with the acquired immunodeficiency syndrome. *N Engl J Med.* 1990;323:109-113.
7. Rogall T, Wolters J, Flohr T, Böttger EC. Towards a phylogeny and definition of species at the molecular level within the genus *Mycobacterium*. *Int J Syst Bacteriol.* 1990;40:323-330.
8. Böttger EC, Teske A, Kirschner P, et al. Disseminated '*Mycobacterium genavense*' infection in patients with AIDS. *Lancet.* 1992;340:76-80.
9. Wald A, Coyle MB, Carlson LC, Thompson RL, Hooton TM. Infection with a fastidious mycobacterium resembling *Mycobacterium simiae* in seven patients with AIDS. *Ann Intern Med.* 1992;117:586-589.
10. Jackson K, Sievers A, Ross BC, Dwyer B. Isolation of a fastidious *Mycobacterium* species from two AIDS patients. *J Clin Microbiol.* 1992;30:2934-2937.
11. Bessesen MT, Shlay J, Stone-Venohr B, Cohn DL, Reves RR. Disseminated *Mycobacterium genavense* infection: clinical and microbiological features and response to therapy. *AIDS.* 1993;7:1357-1361.
12. Nadal D, Caduff R, Kraft R, et al. Invasive infection with *Mycobacterium genavense* in three children with the acquired immunodeficiency syndrome. *Eur J Clin Microbiol Infect Dis.* 1993;12:37-43.
13. Gaynor CD, Clark RA, Koontz FP, Emler S, Hirschel B, Schlesinger S. *Mycobacterium genavense:* disseminated infection in two patients with AIDS. *Clin Infect Dis.* 1994;18:455-457.
14. Tortoli E, Simonetti T, Dionisio D, Meli M. Cultural studies on two isolates of '*Mycobacterium genavense*' from patients with AIDS. *Diagn Microbiol Infect Dis.* 1994;18:7-12.
15. Centers for Disease Control. Revision of the CDC surveillance case definition for acquired immunodeficiency syndrome. *MMWR Morb Mortal Wkly Rep.* 1987;36:3S-15S.
16. Ward DM, Weller R, Bateson MM. 16S rRNA sequences reveal numerous uncultured microorganisms in a natural community. *Nature.* 1990;345:63-65.
17. Edwards U, Rogall T, Blöcker H, Emde M, Böttger EC. Isolation and direct complete nucleotide determination of entire genes: characterization of a gene coding for 16S ribosomal RNA. *Nucleic Acids Res.* 1987;17:7843-7853.
18. Böddinghaus B, Rogall T, Flohr T, Böttger EC, Blöcker H. Detection and identification of mycobacteria by amplification of rRNA. *J Clin Microbiol.* 1990;28:1751-1759.
19. Heald A, Flepp M, Chave JP, et al. Treatment for cerebral toxoplasmosis protects against *Pneumocystis carinii* pneumonia in patients with AIDS: the Swiss HIV Cohort Study. *Ann Intern Med.* 1991;115:760-763.
20. Rothman KJ: *Modern Epidemiology.* Boston, Mass: Little Brown & Co; 1986.
21. Hoop RK, Bottger EC, Ossent P, Salfinger M. Mycobacteriosis due to *Mycobacterium genavense* in six pet birds. *J Clin Microbiol.* 1993;31:990-993.
22. Horsburgh CR Jr, Havlik JA, Ellis DA, et al. Survival of patients with acquired immune deficiency syndrome and disseminated *Mycobacterium avium* complex infection with and without antimycobacterial chemotherapy. *Am Rev Respir Dis.* 1991;144:557-559.

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