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Clarithromycin-Ciprofloxacin-Amikacin for Therapy of *Mycobacterium avium-Mycobacterium intracellulare* Bacteremia in Patients with AIDS

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A combination of clarithromycin, ciprofloxacin, and amikacin for the treatment of *Mycobacterium avium-Mycobacterium intracellulare* bacteremia was evaluated in 12 AIDS patients. Mycobacteremia cleared in all patients by 2 to 8 weeks of treatment, and symptoms resolved. Four patients died; all had negative blood cultures until death, and disseminated *M. avium-M. intracellulare* complex infection was not considered the primary cause of death.

Mycobacterium avium-Mycobacterium intracellulare complex (MAC) infection is one of the most common systemic bacterial infections complicating AIDS. It has been cultured during life from as many as 34% of AIDS patients (8), and according to recent studies, 40 to 50% of all patients with AIDS appear to be infected (6). Moreover, many studies show that disseminated MAC infection is a major cause of illness and makes a substantial contribution to the death of these patients (3).

Early reports on the effect of treatment were discouraging, with persistent bacteremia despite therapy. Recent trials with newer antimicrobial agents have met with only limited success (6, 13, 19), and at present, the most effective therapeutic regimen is unknown.

Clarithromycin, a new macrolide antibiotic structurally related to erythromycin, has recently been found to have, in vitro, favorable activities against MAC isolates, inhibiting 90% of strains at a MIC of 1 to 8 μg/ml (1, 5, 11, 14, 18). It has also been shown to concentrate inside macrophages and tissues (4, 5, 15–17) and to be more acid stable than erythromycin. For these reasons, concentrations lower than the MIC might in vivo be as effective as the higher ones. It is, moreover, possible that clarithromycin, though more active in vitro at a physiologic pH than at an acid pH (18), would remain active for a long period of time in the acid environment of the phagolysosome.

Clinical experience with clarithromycin in MAC-infected AIDS patients is lacking. A prospective, double-blind, placebo-controlled trial in disseminated MAC infection has recently been reported by Dautzenberg et al. (2). In this study MAC disappeared from the blood cultures of seven of eight clarithromycin-treated patients. Five of these patients were cleared of mycobacteria after 2 weeks of treatment, and the other two were cleared after 4 weeks. By contrast, MAC increased in the five patients who received the placebo.

According to some authors, another two compounds, ciprofloxacin and amikacin, have nowadays been recognized to have a good anti-MAC activity both in vitro and in experimental infection (7, 9–11).

We report the results of a pilot study on the value of a triple combination of clarithromycin, ciprofloxacin, and amikacin in the treatment of AIDS patients with persistent MAC bacteremia.

In the period from October 1989 to July 1990 patients admitted to the Infectious Diseases Departments of Vicenza and Pavia Hospitals were included in the study if (i) human immunodeficiency virus antibodies and at least two pretherapy MAC cultures from blood were positive, (ii) symptoms compatible with MAC infection (i.e., fever, night sweats, weight loss, and weakness) were present, (iii) at least 4 weeks of treatment with the triple-drug therapy had been given, and (iv) results were available for two or more cultures from blood (and in some patients also from other sites) drawn at least 4 and then 6 to 8 weeks after the treatment was begun.

All patients received amikacin (7.5 mg/kg of body weight intravenously, two times daily) for 3 weeks, clarithromycin (1,000 mg orally, two times daily), and ciprofloxacin (500 mg orally, three times daily). Ciprofloxacin and clarithromycin were continued indefinitely.

Blood cultures were performed at 2- to 4-week intervals in the first 2 months of treatment and then at 4- to 8-week intervals for 6 to 40 weeks. Patients were considered to have cleared mycobacteremia if at least two consecutive blood cultures were found to be negative.

Clinical response was evaluated by assessing, before and during therapy, the symptoms and signs suggestive of MAC infection.

Adverse reactions were assessed by monitoring both clinical manifestations and laboratory parameters. Complete blood counts and renal and liver tests were performed every 7 days in the first month of treatment and then every month. Audiometric testing could not be performed.

Blood specimens for culture were processed with the Isolator lysis-centrifugation system (Du Pont Co., Wilmington, Del.) as previously described (12). Results were reported as positive or negative; the number of CFU per milliliter of blood was not recorded. MAC was identified by using the Gen-Probe Rapid Diagnostic System for MAC

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TABLE 1. Effect of the triple-drug therapy on MAC bacteremia in AIDS patients

Patient no.	Type[a] (no.) of pretherapy cultures positive for MAC	Wk of therapy with blood culture		Wk of survival on therapy (cause of death)
		Positive	Negative	
1	B (3), BM, F	4	6, 8, 10, 12, 16, 24, 32, 36, 40	44 (bronchopnenmonia)[b]
2	B (2), BM, F	None	4, 6, 8, 12	12 (heart failure)[b]
3	B (2), BM	None	4, 6, 8	24 (heart failure)[c]
4	B (2)	None	4, 6,	28 (*P. carinii* pneumonia)[b]
5	B (2)	None	4, 6, 10	20[d]
6	B (2)	None	4, 8, 12	20[d]
7	B (2)	None	4, 8, 16, 20	23[d]
8	B (2)	4	8, 20	24[d]
9	B (2), F	None	4, 12, 20	25[d]
10	B (2), LN	None	2, 6, 12, 20	23[d]
11	B (2)	None	4, 8, 20	25[d]
12	B (2), LN	None	4, 8, 20	24[d]

[a]B, blood; BM, bone marrow; F, feces, LN, lymph node.
[b]Nonautoptical evidence of mycobacteriosis (see text).
[c]Discontinuance of therapy at the 10th week; blood cultures not done since the 8th week of therapy; fecal cultures positive 2 weeks after therapy was discontinued.
[d]Still alive at the end of the study (December 1990).

(Gen-Probe, Inc., San Diego, Calif.). The results were then confirmed by standard biochemical methods.

The in vitro susceptibility was determined by the agar dilution method (10). Only transparent colony types of clinical isolates were used for the test. Mycobacteria were grown for 7 days in Middlebrook 7H9 broth with 0.5% glycerol and 10% OADC (oleic acid, albumin, glucose, and catalase; Difco Laboratories, Detroit, Mich.). This was then adjusted to achieve the turbidity of a McFarland 0.5 standard. The bacterial suspension, which contained 10 CFU/ml, as determined by triplicate plate counts, was spot inoculated with a Multipoints Inoculator onto Middlebrook 7H10 agar plates supplemented with 0.5% glycerol and 10% OADC containing twofold serial dilutions of the test antibacterial agent. The plates were taped, placed in gas-permeable plastic bags, and incubated in a humidified atmosphere at 35°C for 7 to 21 days.

The antibiotics tested and their sources were as follows: clarithromycin, Abbott Laboratories, North Chicago, Ill.; amikacin, isoniazid, rifampin, and ethambutol, Sigma Chemical Co.; rifapentin, Lepetit Research Center, Gerenzano, Italy; and roxithromycin, Hoechst-Roussel Pharmaceuticals, Inc., Somerville, N.J. Stock solutions were prepared in accordance with the manufacturers' instructions and kept in aliquots at −70°C.

A total of 12 patients (8 males and 4 females; mean age, 29.1 ± 5.3 years; range, 21 to 42 years) met the criteria for evaluation. Seven were intravenous drug abusers, two were homosexual men, and three (two females and one male) were heterosexuals. The AIDS-defining illness was disseminated MAC infection in one, *Pneumocystis carinii* pneumonia in six, esophageal candidiasis in two, neurotoxoplasmosis in one, and wasting syndrome in two. Among the 11 already diagnosed with AIDS, disseminated MAC infection was diagnosed a mean of 5.7 ± 4.5 months later.

As for the symptoms suggesting MAC infection, fever, night sweats, fatigue, and anorexia were present in all patients, hepatomegaly and/or splenomegaly was present in 9, and occasional loose stools or diarrhea was present in 6. The CD4 cell counts at enrollment were 4 to 200 cells/mm^c (mean: 77.1 ± 50.6). Eleven patients had abnormal baseline alkaline phosphatase levels.

Eight patients had previously received, for a period of 2 to 32 weeks (mean: 11.8 weeks), different combinations of antimycobacterial drugs, usually including rifampin, rifapentin, amikacin, clofazimine, isoniazid, and ethambutol. At the start of the clarithromycin-amikacin-ciprofloxacin regimen, any other antimycobacterial treatment had, however, been discontinued for a minimum of 2 weeks.

Patients were given clarithromycin-ciprofloxacin-amikacin for 10 to 44 weeks (see Table 1).

Table 1 shows the pretherapy culture results and the times of positive and negative blood cultures for MAC by week after the triple-drug combination treatment was started. All patients had two or more consecutive negative blood cultures after treatment was begun. The first negative culture was obtained 4 weeks after therapy was started in nine patients and after 2, 6, and 8 weeks of treatment in the remaining three (mean, 4.3 ± 1.4 weeks).

Two patients (Table 1, patients 1 and 8) were found to be positive at the 4-week blood culture. Nine (patient 1) and two (patient 8) subsequent blood cultures, collected over 36 and 16 weeks, respectively, were, however, negative. In one patient (no. 1) the bacterioscopic stool examination 10 weeks after starting therapy was positive for acid-fast bacilli. However, stool culture did not confirm this positive result, and culture of blood drawn on the same day was found to be negative for MAC.

One patient (Table 1, patient 3) had medication stopped by his physician 10 weeks after treatment was started; bacterioscopic examination and culture of feces 2 weeks after therapy was discontinued were positive for MAC. Unfortunately, blood cultures were not available.

Specimens from patient 4, clinically assessable for 28 weeks, could be cultured only twice, 4 and 6 weeks after treatment was started.

Overall, microbiological follow-up (blood cultures) could be continued for 40 weeks in one case, 20 weeks in six cases, 12 weeks in two cases, 10 in one case, and 8 and 6 weeks in one patient each (Table 1).

Of the 12 patients, 4 died, and 3 of these (no. 1, 2, and 4) were autopsied. Well-formed granulomas with necrosis in hematoxylin-eosin staining of bone marrow, liver, spleen, and lymph nodes were not seen. Acid-fast bacilli in smears from the same organs were likewise not observed. For patients 1 and 2, cultures from bone marrow, liver, spleen,

and lymph nodes in Lowenstein-Jensen medium were also done, and they were negative for MAC. All four patients had negative blood cultures until death; disseminated MAC infection was not considered to be the primary cause of death, which appeared to be bronchopneumonia, *P. carinii* pneumonia, and heart failure (one each for the autopsied patients).

As for the clinical response, fever and night sweats resolved in all patients, usually within 2 weeks of commencing triple-drug treatment. Nine of 12 patients also gained approximately 2 kg of weight within the first month of treatment. Symptoms recurred in one patient (no. 3) after discontinuation of therapy.

With regard to adverse reactions, three (25.0%) and two (16.6%) patients complained of abdominal pain and nausea or vomiting, respectively, probably related to therapy. Indeed, these symptoms were relieved by giving clarithromycin (500 mg) every 6 h and ciprofloxacin 3 h after clarithromycin. Moreover, modification of baseline values of liver enzyme was observed in four (33.3%) subjects. Skin rash was not seen. Clinical adverse reactions were mild and transitory, and abnormalities in liver function tests were minimal (less than twice the baseline values in two patients) or mild (more than twice and less than five times the baseline values in two patients). Therapy was therefore continued without modification of the total daily doses.

In vitro antimycobacterial susceptibility tests were performed on 10 isolates. The MICs for 90% of MAC isolates were as follows: amikacin, 16 μg/ml; ciprofloxacin, 8 μg/ml; clarithromycin, 8 μg/ml; roxithromycin, 32 μg/ml; rifampin, >16 μg/ml; rifapentin, 16 μg/ml; isoniazid, >16 μg/ml; and ethambutol, 16 μg/ml.

Overall, the results of our pilot study seem very encouraging. Thus, controlled studies with a larger number of patients should be performed in order to precisely define the role of this therapeutic approach to MAC bacteremia in AIDS patients.

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