Clostridium difficile Isolation in Leukemic Children on Maintenance Cancer Chemotherapy

A Preliminary Study

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Between December 1982 and November 1983, stool specimens from 15 children with acute lymphoblastic leukemia, who were on maintenance cancer chemotherapy, were examined weekly for the presence of *Clostridium difficile* and its toxin. Four out of 15 patients were positive for *C. difficile:* three patients had stool specimens that did not contain toxin, but cultures yielded growth of toxigenic *C. difficile* on only one occasion. The fourth patient, who had a recent history of hospitalization, particularly aggressive cancer chemotherapy, neutropenia, and antibiotic therapy, excreted both *C. difficile* and its toxin for at least 1 month. All children were asymptomatic at the time of positive cultures. This preliminary study reveals a low rate of *C. difficile* colonization in leukemic children on maintenance cancer chemotherapy.

CLOSTRIDIUM DIFFICILE HAS BEEN recognized as a cause of anti-microbial agent-associated colitis or diarrhea in humans and in hamsters.[1-3] Furthermore, it has been reported recently that cancer chemotherapeutic agents also may have a causative role in *C. difficile*-induced disease.[4] A small number of patients treated with chemotherapy for malignancy who subsequently developed diarrhea with both *C. difficile* and its toxin present in their stools have been reported.[5-7] To date, however, no study has been attempted to assess their role in patients on maintenance cancer chemotherapy. Consequently, we studied weekly stool specimens obtained over a 12-month period from 15 children with acute lymphoblastic leukemia, who were on maintenance cancer chemotherapy, for the presence of *C. difficile* and its toxin.

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Subjects and Methods

Patient Population

Between December 1982 and November 1983, stool specimens were collected weekly from 15 children with acute lymphoblastic leukemia who were seen as outpatients at the Pediatric Clinic, University of Rome, and screened for the presence of *C. difficile* and its toxin. Informed written consent was obtained from parents whose children entered the study. Nine subjects were male and six were female (Fig. 1). Their mean age was 8 years (range 4–14 years). Fourteen children had common lymphoblastic leu-

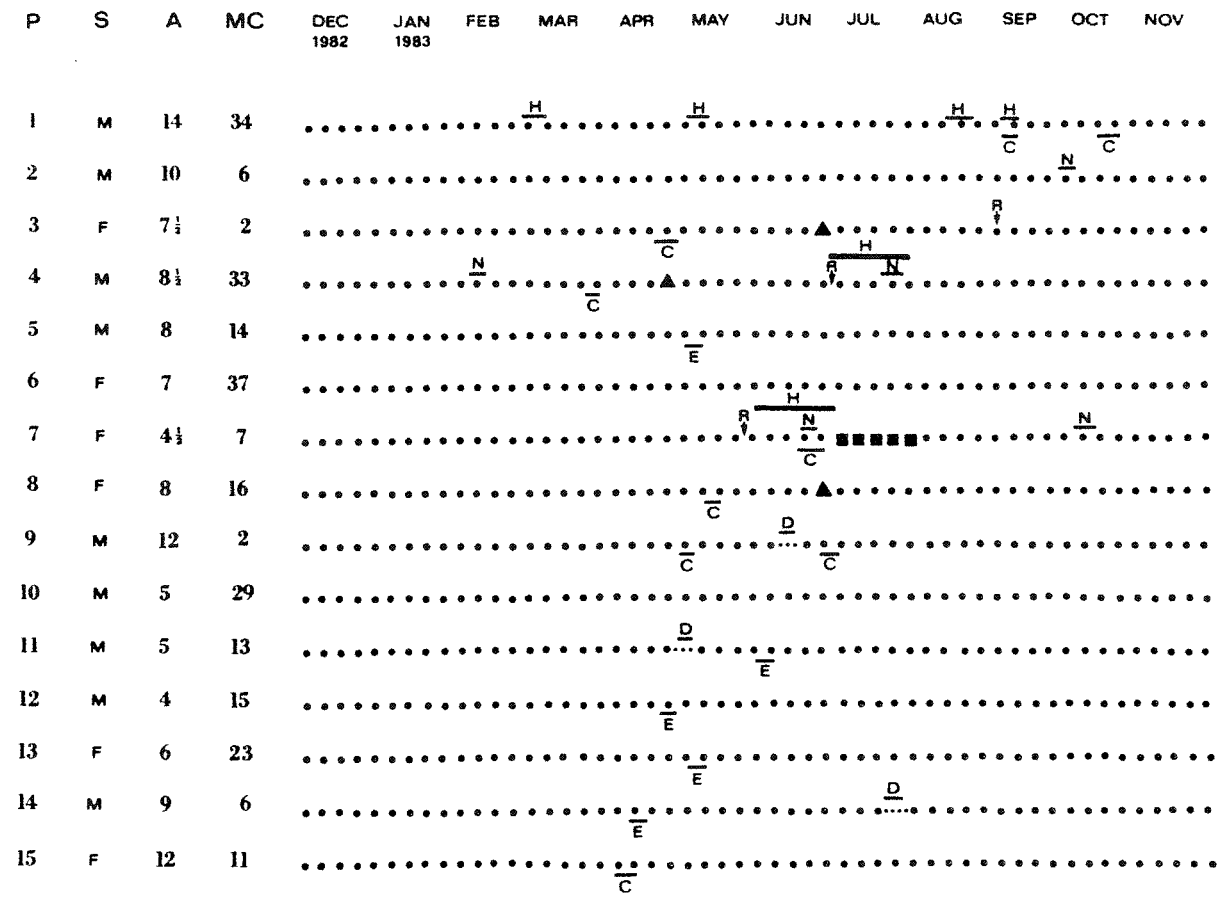

FIG. 1. Clinical and laboratory data throughout the 1-year study of *Clostridium difficile* in leukemic children. P: Patient; S: Sex; A: Age in years (prior to the onset of study); MC: Number of months of maintenance cancer chemotherapy (prior to the onset of study); •: Stool specimens collected weekly between December 1982 and November 1983; ·: Daily fecal specimens obtained throughout the entire diarrheic episode; H: Hospitalization*; N: Neutropenia*; R: Relapse; C: Treatment with cephalosporins*; E: Treatment with erythromycin*; D: Diarrhea*; ▲: *C. difficile* isolated/toxin negative; ■: *C. difficile* isolated/toxin present; *The length of bar indicates the duration of the event.

kemia and one patient (7) had T-cell acute lymphoblastic leukemia.

The study population at the outset was in remission and on maintenance cancer chemotherapy, which included the following regimens: methotrexate (MTX), vincristine (VCR), 6-mercaptopurine (6-MP), and prednisone (PDN) (patients 2, 3, 5, 7, 8, and 10 through 15); thioguanine (TG), cyclophosphamide (CPM), daunomycin (DNM), bischlorethyl nitrosourea (BCNU), MTX, VCR, and cytosine arabinoside (ARA-C) (patients 4 and 9); ARA-C, MTX, VCR, and PDN (patient 1); and MTX, VCR, ARA-C, adriamycin (ADR), 6 MP, and PDN (patient 6). The median duration of remission was 16 months (range 2–37 months). Patients 3, 4, and 7 experiencing central nervous system relapse were reinduced with VCR, PDN, MTX, L-asparaginase (LASP), and ARA-C. MTX and ARA-C were given intrathecally.

During the study, 12 patients were treated with antibiotics for 3 to 10 days because of upper respiratory tract infection and/or fever: 7 subjects received cephalosporins and five received erythromycin. Over the 1-year period, three children developed diarrhea that was mild, self-limited, and unrelated to antibiotic exposure. Daily fecal specimens were obtained in all three cases throughout the entire diarrheal episode.

Bacteriological Methods

Fresh stool samples were evaluated for *C. difficile* using the selective cycloserine-cefoxitin-fructose agar, which was prepared according to George *et al.*,[8] and incubated in an anaerobic atmosphere. Bacterial isolates were identified by standard laboratory methods, using biochemical reactions and gas-liquid chromatography. Isolates of *C. difficile* were tested for cytotoxin *in vitro* production, as described by Chang *et al.*[9]

Concomitant with isolation techniques, stools were assessed for *C. difficile* toxin as described previously.[9] The *C. difficile* anti-toxin used in this study was kindly supplied by Tracy Wilkins of the Virginia Polytechnic Institute and State University, Blacksburg, VA. Medical records of all study patients were reviewed to correlate clinical and laboratory data.

Results

Throughout 1 year, a total of 789 samples of feces were processed. *C. difficile* was recovered from the stools of four children (patients 3, 4, 7, and 8) who had received cephalosporins within the previous 8 weeks (Fig. 1). All *C. difficile* isolates had the ability to produce toxin *in vitro*. At the time of positive cultures, none of the patients had diarrhea or any inflammatory bowel disease.

Patients 3, 4, and 8 did not have toxin in their stools, and *C. difficile* could not be demonstrated in the successive fecal samples. In the fourth patient (case 7), who had been hospitalized for 30 days because of central nervous system relapse and had received reinduction therapy leading to neutropenia (neutrophils = 650/μl), both *C. difficile* and toxin were found in the stools 2 days after discharge from the hospital and 4 days following the discontinuation of antibiotic treatment. This patient continued to be positive for *C. difficile* and toxin for at least 1 month. The toxin titer was 1 to 400 in all stool specimens and dropped to 1 to 40 in the last positive assay. Stool cultures performed on patients 9, 11, and 14, who were affected by diarrhea, were repeatedly negative.

Discussion

The results of our preliminary study, centered mainly on an outpatient population, indicate that leukemic children on maintenance cancer chemotherapy occasionally harbor *C. difficile*. Of 15 children followed weekly for 1 year, only four were positive for *C. difficile:* three had stool specimens that did not contain toxin, but cultures yielded growth of toxigenic *C. difficile* on only one occasion. The fourth patient excreted *C. difficile* and its toxin for 4 weeks (patient 7). All four subjects had received cephalosporins prior to positive cultures.

George and co-workers isolated *C. difficile* and fecal toxin from asymptomatic patients receiving cephalosporins, but they failed to specify the time that elapsed between drug administration and *C. difficile* isolation.[3] In this study, on the basis of the interval between prior antibiotic exposure and *C. difficile* recovery, this association is unlikely in cases 3, 4, and 8.

Our data, which were based on patients who had only limited hospitalization and antibiotic exposure, and were treated with doses of chemotherapy of moderate intensity, do not permit conclusions in nonleukemic cancer patients or in patients receiving more intensive doses of therapy or requiring more in-hospital therapy. However, although a small number of children was monitored, many stool samples were processed over a 1-year period. Therefore, we suggest that *C. difficile* is not a major problem in leukemic children on maintenance cancer chemotherapy. In fact, our bacteriologic findings are indicative of a low rate of *C. difficile* colonization in this population.

Moreover, *C. difficile* and its toxin did not play a pathogenic role in our patients, since none of the subjects developed symptoms or serious consequences. Patient 7 had been hospitalized previously for 30 days for reinduction chemotherapy following central nervous system relapse, and had received antibiotics because of fever and neutropenia. He was 5 years old, an unusual age for such a finding,[10] and harbored both *C. difficile* and toxin for 1 month without developing symptoms. This patient's course suggests the need for additional factors to explain *C. difficile*-related disease.

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CLINICAL NOTE

Diphtheria-Tetanus-Pertussis Vaccine Shortage—United States*

Major changes have occurred in the pattern of manufacture and distribution of diphtheria-tetanus-pertussis (DTP) vaccine in the United States. Two of the three U.S. commercial manufacturers (Wyeth and Connaught, Inc.) have stopped distribution of their products. Thus, only one manufacturer (Lederle) now markets DTP vaccine in the United States. Lederle has been increasing its population and expanding its facilities to meet current needs. Careful monitoring of supplies and production schedules previously indicated that national supplies would be adequate. However, some recent lots of Lederle DTP vaccine have failed to meet the manufacturer's requirements for release. Production and testing of this three-component vaccine is complex and requires several months. Comparison of available stocks and the quantity of DTP vaccine now being distributed with the usual national utilization of DTP vaccine indicates that, if current use pattens continue, beginning in January 1985, supplies of DTP vaccine will be very limited, and some areas may be without DTP vaccine. This situation may continue through most of 1985.

To minimize the health impact of this shortage, two major options exist—to reduce the amount of vaccine given in a particular dose and to postpone one or more doses. Because it is impossible to predict the degree of protection conferred by partial doses, this option is not recommended.[1] Consequently, consideration has been given to the possibility of postponing one or more doses of the current immunization schedule, which calls for the administration of DTP vaccine at 2, 4, 6, and 18 months of age, with a fifth dose at 4 to 6 years of age.

With pertussis, there is a significant risk of infection in infancy and early childhood, with 2463 cases reported in 1983 (51% of them among infants under 1 year old). Additionally, infants are more likely to suffer complications or death from pertussis than are older children. Consequently, it is critical to continue providing protection against pertussis to infants. The first three doses of DTP vaccine provide protection against pertussis in 70 to 90% of recipients and immunity to diphtheria and tetanus in over 90 percent of recipients.[2-4] The doses given at 18 months and at 4 to 6 years of age enhance protection through the preschool and early school years, respectively.

Taking all these factors into account, interim postponement of the doses of DTP vaccine given at 18 months and at 4 to 6 years of age could achieve substantial savings in the rate of DTP vaccine use, while still protecting those at greatest risk of these diseases.

After consultation with members of the Immunization Practices Advisory Committee and the Committee on Infectious Diseases of the American Academy of Pediatrics, the following interim recommendations are made:

> Effective immediately, all health-care providers should postpone administration of the DTP vaccine doses usually given at 18 months and 4 to 6 years of age (fourth and fifth doses) until greater supplies are available. When adequate DTP vaccine becomes available, steps should be taken to recall all children under 7 years of age who miss these doses for remedial immunization.

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* MMWR December 14, 1984;33:695-6.