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Trimethoprim and Rifampin in Combination for Chemoprophylaxis of Household Contacts of Patients with Invasive Infections Due to *Haemophilus influenzae* Type b

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We compared the effectiveness of rifampin-trimethoprim in fixed combination (3.75:1) to rifampin alone in the eradication of *Haemophilus influenzae* type b carriage among contacts of patients with invasive infection caused by this organism. The study population was composed of 127 index patients and 620 contacts. Twenty-six percent of contacts were colonized. Rifampin-trimethoprim eradicated carriage in 77.6% of contacts (71.1% in contacts <5 years, 84.2% in contacts ≥5 years) whereas rifampin eradicated carriage in 69.9% of contacts (56.4% in contacts <5 years, 81.8% in contacts ≥5 years). A single isolate resistant to rifampin and rifampin-trimethoprim was encountered. The eradication rate achieved with this regimen of rifampin-trimethoprim was too low to recommend its routine use. However, a higher dose or longer course might merit clinical trial.

The high secondary attack rate documented among contacts (5) of patients with invasive *Haemophilus influenzae* type b (Hib) disease has stimulated efforts to identify an antibiotic regimen which might interrupt transmission of Hib among those at risk. To date, the most promising chemoprophylactic agent has been rifampin. However, the isolation of *H. influenzae* strains resistant to this agent (10, 11) has raised concern that its effectiveness might be compromised by widespread use. Recently, in vitro synergy has been demonstrated for trimethoprim and rifampin in combination against *H. influenzae* (1, 7). In addition, the use of two antimicrobial agents together might diminish the likelihood of isolating resistant mutants (12). Therefore, we conducted a multicenter study to evaluate the effectiveness of rifampin and trimethoprim in combination in the eradication of Hib carriage among contacts of patients with invasive Hib infections.

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MATERIALS AND METHODS

The design of this study was similar to that in a previous study of ours in which rifampin and placebo were compared (4). Briefly, all patients admitted to Charity Hospital, New Orleans, Children's Hospital Medical Center, Boston, or Children's Hospital, Denver, with Hib bacteremia, meningitis, arthritis, epiglottitis, pericarditis, cellulitis, or pneumonia, diagnosed as previously described, were offered participation in the study. An adult family member was contacted, and a roster of household contacts (4) was compiled. A patient contact group (PCG) consisted of individuals who normally resided in the home of the index patient or who spent 4 h or more there per day for 5 of the 7 days before hospitalization of the index case. In formed consent was obtained for each contact; patient contacts were randomized to receive either rifampin in a dose of 10 mg/kg (maximum dose, 600 mg) or rifampin-trimethoprim in a dose of 10 mg of rifampin per kg (maximum dose, 600 mg) and 2.67 mg of trimethoprim per kg (maximum dose, 160 mg) twice daily for 2 days. Rifampin-trimethoprim was supplied as sugar-coated tablets in a fixed ratio of 3.75:1. For pediatric dosing, rifampin-trimethoprim tablets were ground in an electric dry-food grinder, and a preweighed dose of powder was dispensed in cellophane packets. Rifampin powder (Rifadin; Dow Pharmaceuticals, Indianapolis, Ind.) was dispensed in preweighed portions. Parents were instructed to mix the powder with a small amount of applesauce. All contacts of a single patient, both children and adults, received the same medication. The index patient was treated with the identical regimen at discharge. Pregnant women did not receive either drug. Throat cultures, obtained before antibiotic therapy and 10 days after the initial visit, were plated immediately on

antiserum agar (9). At the follow-up visit 10 days later, contacts were asked whether they had complied with the prescribed regimen and were individually surveyed with a questionnaire (4) for toxicity associated with drug administration. Identification and storage of Hib strains were performed as previously described (4).

Susceptibility testing for rifampin and rifampin-trimethoprim at the ratio contained in the tablet was performed as follows. Rifampin (supplied by Dow Chemical Co.) was dissolved in methanol and diluted with distilled water. Trimethoprim (Sigma Chemical Co., St. Louis, Mo.) was dissolved in 0.1 N HCl and diluted with distilled water. All assays were performed in microtiter trays. Rifampin concentrations in plates, used to determine minimal inhibitory concentrations (MICs), ranged from 30.0 to 0.03 μg/ml. Rifampin-trimethoprim concentrations ranged from 30.0-8.0 μg/ml to 0.03-0.01 μg/ml. Mueller-Hinton broth (0.05 ml) supplemented with hemin (Eastman Kodak Co., Rochester, N.Y.; final concentration, 1 mg/ml), β-NAD (Sigma; final concentration, 1 μg/ml), and thymidine phosphorylase (Burroughs Wellcome Co., Research Triangle Park, N.C.; final concentration, 0.05 U/ml) and containing the appropriate antibiotic concentration was dispensed into each well. Inocula were prepared by adjusting a mid-log-phase broth culture of 5×10^8 CFU/ml to ca. 5×10^5 CFU/ml by dilution in the supplemented Mueller-Hinton broth. Each well of the microdilution plate, used to determine MICs, was inoculated with 0.05 ml of this suspension. Thus, the final inoculum was ca. 10^4 CFU. Inoculated plates were incubated at 37°C for 18 h in 5% CO_2.

The MIC was defined as the lowest concentration of antibiotic which inhibited growth. Minimal bactericidal concentrations (MBCs) were determined by subculturing from the microdilution plates with a 10-μl Eppendorf pipette (Brinkmann Instruments, Inc., Westbury, N.Y.). Samples from each clear well were plated on brain heart infusion agar supplemented with hemin (final concentration, 1 mg/ml) and β-NAD (final concentration, 1 μg/ml) and incubated overnight. Thus, the MBC was defined as the lowest antibiotic concentration from which no viable organisms could be recovered (>99.9% kill). Isolates tested included the pre- and posttreatment isolates from 40 of 42 contacts who remained colonized with Hib after prophylaxis.

The X^2 test was used to compare the carriage eradication rates of the rifampin and rifampin-trimethoprim groups. A log linear model was used to test for the distribution of Hib carriage by age and for the infectious syndrome of the index patient. The X^2 test was used to compare the incidence of side effects between the drug groups. A paired t test was used to compare the MICs and MBCs of the drugs for pre- and posttreatment Hib isolates in contacts who remained colonized after prophylaxis.

RESULTS

Index patients and patient contact groups. During the study period, 127 index patients (New Orleans, 43; Denver, 45; Boston, 39) were hospitalized during the study period. At the initial interview, 620 household contacts were identified. The mean age of the index patients was 17.4 months. Of the 127 index patients, 71% had meningitis, 8% had epiglottitis, and 21% had other Hib syndromes; 58% were male; and 15% attended a day-care center.

A PCG contained a mean of 5.1 individuals. Of the PCGs, 63% included at least one individual <5 years of age, in addition to the index case.

Carriage among contacts. Of 620 patient contacts, 161 (26.0%) were initially colonized with Hib. The distribution of colonized contacts by age resembled that observed previously (4). Hib colonization in children ≤9 years of age was documented in 116 of 220 individuals (52.7%), whereas 45 of 400 individuals (11.3%) >9 years were colonized ($P < 0.0001$; X^2). In contacts <5 years of age, the carriage rate varied significantly ($P < .05$) with the infectious syndrome of the index case: in contacts <5 years of age, 62% were colonized when the index patient had meningitis, 18% were colonized when the infectious syndrome of the index patient was epiglottitis, and 32% were colonized when the index patient had other Hib syndromes.

Follow-up and compliance. Of the 620 PCG members, 559 (90.1%) returned 10 days after the initial visit for follow-up culture. A study drug was not prescribed for nine of these individuals who were pregnant. Of the 550 members who returned and were eligible for prophylaxis, 524 (95.3%) reported full compliance with the prescribed regimen.

Eradication and acquisition of Hib carriage. Of the 161 contact-carriers identified initially, 152 (94.4%) returned 10 days later for follow-up. Eight of these individuals were excluded from the calculation of eradication rates; rifampin was contraindicated in one, and seven took three or fewer doses of rifampin. Thus, 144 contact-carriers reported full compliance and returned for follow-up. We also studied 15 compliant carriers who had requested inclusion in the study despite insufficient contact with the index patient to qualify as an authentic PCG member. A prophylactic regimen identical to that of the bona fide PCG members was prescribed for these individuals. Data regarding carriage eradication in the 159 carriers are shown in Table 1. The rate of acquisition of Hib carriage between the time of the initial culture and the follow-up visit 10 days later did not differ significantly when all contacts (2.4% rifampin versus 2.5% rifampin-trimethoprim), contacts ≥5 years of age (0.7% rifampin versus 0.6% rifampin-trimethoprim), and contacts <5 years of age (12.0% rifampin versus 12.5% rifampin-trimethoprim) were compared.

Toxicity. The incidence of side effects reported by contacts receiving rifampin was compared with that reported by rifampin-trimethoprim re-

TABLE 1. Results of day 10 follow-up cultures among 144 compliant contact-carriers (plus 15 occasional contacts)

Population	Treatment	No. of carriers	% Conversion (positive to negative)[a]
All compliers	Rifampin-trimethoprim	76	77.6
	Rifampin	83	69.9
<5 yr	Rifampin-trimethoprim	38	71.1
	Rifampin	39	56.4
≥5 yr	Rifampin-trimethoprim	38	84.2
	Rifampin	44	81.8

[a] For each population, percent conversion in rifampin-trimethoprim-treated contacts was compared with percent conversion in rifampin-trimethoprim-treated contacts, using the two-tailed test. P = not significant in all populations.

cipients. No significant differences were observed in the toxicity of the two regimens (Table 2).

Susceptibility testing. The MICs and MBCs for pretreatment isolates from 40 of 42 contacts who remained colonized after prophylaxis with either regimen were compared with those in the corresponding posttreatment isolates. The mean MIC of rifampin for 40 pretreatment isolates was 0.34 μg/ml (range, 0.06 to 1.88 μg/ml); after therapy, the mean MIC was 0.39 μg/ml (range, 0.06 to 1.88 μg/ml). For rifampin-trimethoprim, the pretreatment mean MIC averaged 0.33-0.09 μg/ml (range, 0.06-0.02 to 0.94-0.25 μg/ml), whereas the mean MIC after therapy was 0.35-0.09 μg/ml (range, 0.03-0.01 to 0.94-0.25 μg/ml). The MICs of rifampin and rifampin-trimethoprim for isolates of patients who remained colonized after prophylaxis did not differ with the prescribed regimen. In 37 of 40 strains tested, the MIC and MBC of rifampin-trimethoprim were identical. In three strains, the MBC (0.94-0.25 μg/ml) exceeded the MIC (0.12-0.04 μg/ml) by three dilutions. Synergy at a ratio of rifampin to trimethoprim of 3.75:1 could not be inferred since the MIC of rifampin when tested in combination with trimethoprim was never more than a single dilution lower than the MIC of rifampin when tested alone.

One isolate of *H. influenzae* resistant to rifampin was encountered in an individual who did not satisfy the contact criteria but who took rifampin after isolation of Hib from the pharynx. Before treatment the MIC and MBC of rifampin had been 0.47 μg/ml, whereas the MIC and MBC of rifampin-trimethoprim were 0.23-0.07 and 0.47-0.13 μg/ml, respectively. This individual remained a carrier after rifampin therapy and then was treated with rifampin-trimethoprim. However, Hib was again isolated from the oropharynx 10 days after therapy. The MIC and MBC of rifampin for this isolate were both 3.75 μg/ml, whereas the MIC and MBC of rifampin-trimethoprim were 1.88-0.5 and 7.5-2 μg/ml, respectively.

DISCUSSION

We previously reported our experience with rifampin at the dose and regimen currently recommended for household contacts of patients with serious meningococcal infections (20 mg/kg per day twice daily for 2 days; maximum dose 1,200 mg per day) (6). Overall, rifampin eradicated carriage more effectively than did placebo in household contacts. However, in children <5 years of age, the rifampin eradication rate (47.8%) did not significantly exceed that observed with placebo (28.6%).

The combination of rifampin-trimethoprim was not significantly better than rifampin alone among all household contacts (Table 1). In children <5 years of age, the eradication rate with rifampin-trimethoprim (71.1%) was higher than that observed with rifampin alone (56.4%), but this difference was not significant ($X^2 = 1.33$; $P = 0.18$).

We had hypothesized that rifampin-trimethoprim might offer a distinct advantage over rifampin alone. Alvarez et al. (S. Alvarez, A. De-

TABLE 2. Incidence of side effects reported by contacts who took one or more doses of rifampin or rifampin-trimethoprim (n = 538)

Complaint	% Individuals who took[a]: Rifampin (n = 242)	Rifampin-trimethoprim (n = 296)
Drowsiness	0.4	2.0
Fatigue	0.0	1.0
Headache	0.8	2.4
Rash	0.4	1.4
Nausea	2.9	2.4
Fever	0.0	0.7
Vomiting	1.7	1.7
Dizziness	1.2	1.4
Diarrhea	2.1	0.3
Sore mouth	0.4	1.4
Cramps	0.8	0.3
Abdominal pain	1.7	2.0
Muscle pain	0.0	0.3
Itching	0.4	0.3
Red urine	96.3	98.7
Other	3.3	1.7

[a] No significant differences were noted between the treatment groups for any side effect by the χ^2 test. The side-effect survey was inadvertantly omitted for 12 contacts.

maria, J. O. Klein, and W. R. McCabe, Program Abstr. Intersci. Conf. Antimicrob. Agents Chemother. 19th, Boston, Mass., abstr. no. 359, 1979) studied 81 clinical *H. influenzae* isolates. With trimethoprim and rifampin in a 1:1 ratio, synergy was documented in 44 of 81 strains (54%). However, when the rifampin-trimethoprim ratio was 7:2, synergy was documented in only 10 strains. McDougal and Thornsberry (7) studied 30 strains of *H. influenzae* (rifampin-trimethoprim ratio, 1:1) and found synergy in 29 strains when bacterial killing was the endpoint. Zweighaft and McCracken (T. R. Zweighaft and G. H. McCracken, Abstr. Annu. Meet. Am. Soc. Microbiol. 1981, A4, p. 1) studied 11 Hib isolates from blood or cerebrospinal fluid and documented synergy in 6 isolates and an additive effect in 4 isolates. Berti et al. (1) documented synergy between rifampin and trimethoprim against 17 strains of *H. influenzae* but only when the trimethoprim concentration employed exceeded the rifampin concentration by at least fourfold. We did not systematically evaluate synergy. However, in our studies the MIC of rifampin in combination with trimethoprim was not lower than the MIC of rifampin alone in all strains tested.

Others have suggested that the combination of rifampin and trimethoprim might prevent the emergence of rifampin-resistant strains of *H. influenzae*. Mutation to rifampin resistance occurs readily in vitro (8, 12). Moreover, failure of rifampin prophylaxis has been associated with isolation of resistant strains of *H. influenzae* (10, 11). Importantly, the in vitro emergence of rifampin-resistant mutants was prevented when trimethoprim was combined with rifampin (12).

In our study, rifampin and subsequent rifampin-trimethoprim prophylaxis failed in an individual in association with isolation of an Hib strain resistant to rifampin. The explanation for failure of eradication in the 41 other individuals who remained colonized after therapy is not known. Problems in during administration were encountered very rarely. With the exception of transient discoloration of urine and menstrual-like bleeding reported by an occasional rifampin recipient taking birth control pills, little toxicity was encountered.

Our data confirm that the 2-day rifampin regimen we employed does not dependably eradicate carriage in children <5 years of age. In addition, administration of rifampin-trimethoprim for 2 days failed to eradicate carriage in 28.9% of children <5 years of age. It is possible that in some failures, the organism was successfully eradicated but the carrier was recolonized during the 10-day study period. Since the goal of prophylaxis is dependable elimination of the reservoir of Hib carriage surrounding the index patient, the failure rate we observed is unacceptably high. Whether a regimen that dependably eradicates the carrier state may be presumed to also reduce secondary invasive disease remains unknown. Nevertheless, if the currently recommended (2, 3) regimen of rifampin in a dose of 20 mg/kg (maximum dose, 600 mg) once daily for 4 days is associated with frequent isolation of resistant strains, rifampin-trimethoprim at a higher dosage or for a longer period of time might merit clinical trial.

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