ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, July 1981, p. 81-87
0066-4804/81/070081-07$02.00/0

Vol. 20, No. 1

Trimethoprim-Sulfamethoxazole Therapy of Experimental *Escherichia coli* Meningitis in Rabbits

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Received 29 December 1980/Accepted 16 April 1981

We used two strains of ampicillin-susceptible *Escherichia coli* to produce meningitis in rabbits and utilized these models (i) to compare the killing effects of parenteral trimethoprim-sulfamethoxazole (TMP-SMZ) and ampicillin on *E. coli* in cerebrospinal fluid after 8 h of treatment and (ii) to measure the penetration of TMP-SMZ and ampicillin into cerebrospinal fluid and the brain. At 16 h after intracisternal inoculation with a test strain, rabbits were treated with TMP (6 mg/kg per h) and SMZ (30 mg/kg per h), ampicillin (40 mg/kg per h), or saline intravenously for 8 h. TMP-SMZ levels were measured by high-pressure liquid chromatography, and ampicillin levels were measured by microbiological assay. Mean ± standard deviation concentrations of TMP, SMZ, and ampicillin in cerebrospinal fluid (mean percent penetration) at the completion of 8 h of therapy were 0.80 ± 0.41 (18%), 15.7 ± 12.1 (27.2%), and 2.6 ± 1.7 (8.9%) μg/ml, respectively. TMP, SMZ, and ampicillin levels in brain homogenate after 8 h of therapy were 0.23 ± 0.07 (6.6%), 3.31 ± 3.3 (5.5%), and 0.6 ± 4.53 (1.9%) μg/g, respectively. TMP-SMZ infusion for 8 h produced a significant reduction in mean bacterial counts in cerebrospinal fluid in both models of meningitis compared with saline controls. The decrease in mean bacterial counts with TMP-SMZ therapy was equivalent to that produced by ampicillin.

Neonatal meningitis due to aerobic, gram-negative bacilli continues to cause significant morbidity and mortality whether or not parenteral antibiotics are supplemented by intrathecal or intraventricular therapy (15). Ampicillin with or without an aminoglycoside is currently recommended for therapy of coliform meningitis. But an increasing number of gram-negative bacilli isolated from neonates with meningitis, including *Escherichia coli,* are resistant to ampicillin (15). Thus, there is a need to evaluate alternative antimicrobial agents for their effectiveness.

One such agent is trimethoprim-sulfamethoxazole (TMP-SMZ). This antibiotic combination has excellent in vitro activity against aerobic, gram-negative bacilli (1, 2). At present there are limited data concerning the use of TMP-SMZ in the therapy of either experimental or clinical bacterial meningitis (7, 10, 16, 19, 22, 23).

Kirwan (10) reported the successful treatment of one patient with meningitis given oral TMP-SMZ, and measured blood and cerebrospinal fluid (CSF) levels of TMP and SMZ. There have also been several case reports (7, 16, 22) which describe the successful treatment of meningitis with oral TMP-SMZ, but there was no documentation of blood or CSF levels. Sabel and Brandberg (23) reported the successful treatment with parenteral TMP-SMZ of eight of nine infants with meningitis who were considered therapeutic failures. Finally, Perfect et al. (19) have recently reported the treatment of experimental *Haemophilus influenzae* type b meningitis in rabbits with parenteral TMP-SMZ. They found that TMP-SMZ was as effective as ampicillin and chloramphenicol in the treatment of this experimental infection. These uncontrolled clinical reports and the animal study lend impetus to further evaluation of parenteral TMP-SMZ in the treatment of bacterial meningitis.

We compared intravenous TMP-SMZ and ampicillin therapy of experimental *E. coli* meningitis in rabbits. Our goals were: (i) to measure the penetration of TMP-SMZ and ampicillin into the CSF and brain and (ii) to compare the bactericidal effects of TMP-SMZ and ampicillin in the CSF after 8 h of treatment.

(This study was presented in part at the 20th Interscience Conference on Antimicrobial Agents and Chemotherapy, New Orleans, September, 1980.).

MATERIALS AND METHODS

Organisms. The two strains of *E. coli,* C7K1 (EC7)

and C13NK1 (EC13), used for these studies were isolated from neonates with meningitis and kindly provided by George McCracken, University of Texas Health Science Center, Dallas. The organisms were maintained on Mueller-Hinton (MH) agar (Difco Laboratories) at room temperature and subcultured weekly.

Drugs for in vitro studies. TMP (lot no. 089) and SMZ (lot no. 709115) were kindly supplied by Hoffmann-La Roche, Inc., Nutley, N.J. TMP and SMZ were solubilized by a method previously described (18). TMP was dissolved in 1 ml of methanol; MH broth was added to yield a final concentration of 20 μg/ml. SMZ was dissolved in 0.3 ml of 0.1 N NaOH; MH broth was added to yield a final concentration of 400 μg/ml. These stock solutions were made every 2 weeks and stored at −20°C. Ampicillin trihydrate was donated by Bristol Laboratories, Inc., Syracuse, N.Y., and was made fresh on the day in vitro tests were performed.

In vitro studies. Minimum inhibitory concentrations (MICs) and minimum bactericidal concentrations (MBCs) of ampicillin and TMP-SMZ were determined by a twofold tube dilution technique in MH broth (27). TMP-SMZ was kept at a fixed concentration ratio of 1:20 (wt/wt).

The inoculum was a dilution of an overnight growth of the test strain in MH broth adjusted by nephelometry (Coleman Junior 6A Spectrophotometer, Coleman Instruments, Maywood, Ill.) to contain 10^5 colony-forming units (CFU) per ml of broth. Thymidine phosphorylase (0.1 IU/ml; Burroughs-Wellcome Co., Research Triangle Park, N.C.) was added to remove any thymidine that may have been present in the broth and that could counteract the antibiotic effects of TMP and SMZ (5).

The MIC was defined as the lowest concentration of drug(s) which inhibited visible growth after 18 h of incubation at 37°C. The MBC was defined as the concentration of drug(s) in the first clear tube which gave no growth after subculture of 0.1 ml onto MH agar and incubation for 18 h.

Experimental model. After overnight growth on MH agar, the test strain was suspended in phosphate-buffered saline and adjusted by nephelometry to obtain an infecting inoculum of approximately 2×10^5 CFU/ml. Sixty-one male and female adult New Zealand white rabbits weighing 1.7 to 3.0 kg were inoculated intracisternally with *E. coli* as previously described (4). Briefly, after obtaining adequate anesthesia with an intravenous infusion of 30 mg of sodium pentobarbital (Abbott Laboratories, Chicago, Ill.) per kg, a cisternal puncture was performed with a 20-gauge needle attached to a 1.0-ml tuberculin syringe. CSF (0.5 ml) was withdrawn, and 0.5 ml of the test strain was injected. The needle was removed, and the animal was returned to its cage.

In a previous study (4) the course of untreated rabbits with EC13 meningitis was determined. An infecting inoculum of 4×10^5 CFU of EC13 produced a uniformly fatal infection in untreated rabbits with a mean survival time of 61 h.

Study design. At 16 h after inoculation 34 rabbits were begun on therapy with TMP-SMZ or ampicillin. Ten rabbits with EC7 meningitis and eight rabbits with EC13 meningitis were treated for 8 h with TMP (6 mg/kg per h) and SMZ (30 mg/kg per h); nine rabbits with EC7 meningitis and seven rabbits with EC13 meningitis were treated for 8 h with ampicillin (40 mg/kg per h). No loading doses were given. The doses of TMP-SMZ were chosen because they produced serum and CSF levels in uninfected rabbits that were similar to those reported in humans (9, 10, 26).

TMP-SMZ was compounded with 10% ethyl alcohol and 1% benzyl alcohol in 5-ml vials and kindly supplied by Hoffmann-La Roche, Inc. TMP-SMZ was diluted with 5% dextrose in water and delivered by continuous intravenous infusion via a peripheral ear vein by using a Harvard infusion-withdrawal pump (model 940; Harvard Apparatus, Millis, Mass.) at a rate of 21.5 ml/h. At this rate no crystallization occurred. Ampicillin was diluted in phosphate-buffered saline and administered by continuous intravenous infusion. Rabbits were restrained by physical means during the infusion, thereby avoiding the negative effects of barbiturate on choroidal CSF production (6).

A second group of 16 animals, 7 with EC7 meningitis and 9 with EC13 meningitis, received a saline infusion for 8 h and served as infected, untreated controls.

A third group, consisting of two uninfected animals, received TMP-SMZ intravenously for 8 h at the same doses previously noted and served as uninfected controls.

A fourth group of nine rabbits was used to determine the course of untreated EC7 meningitis. These untreated rabbits were followed for a maximum of 96 h post-inoculation or until they died. Cisternal punctures were done on all animals that died. Quantitation of bacteria in CSF was performed on rabbits dying less than 18 h after inoculation. Cultures were done on rabbits dying more than 18 hours after inoculation, but no quantitation was performed because cisternal punctures were not usually done until several hours after death. These cultures are therefore reported only as positive or negative.

Before the initiation of therapy, a cisternal tap was performed, and 0.2 ml of CSF was obtained for quantitative culture. At the end of the 8-h infusion, animals were sacrificed by barbiturate overdose. Blood was obtained by heart puncture, and CSF was obtained by cisternal tap. Samples used for determination of antimicrobial concentration were stored at −20°C until assays were performed. A 0.2-ml sample of CSF was used for quantitation of bacteria. The brain was removed immediately after sacrifice by a technique previously described (3). Briefly, after a midline incision was made over the skull and skin was retracted, multiple burr holes were placed, and a skull flap was removed. The brain was removed in toto, washed once in phosphate-buffered saline, quick-frozen in an alcohol bath, and stored at −20°C. Samples from uninfected rabbits were taken in a similar fashion.

Quantitation of bacteria. The numbers of *E. coli* in CSF were determined by plating 0.1-ml samples of undiluted or serial 10-fold dilutions of spinal fluid on MH agar. Colony counts were determined after incubation at 37°C for 24 h and expressed as CFU per milliliter of CSF.

Ampicillin assay. Ampicillin concentrations in serum, CSF, and brain were determined by means of an

agar-well diffusion technique with *Bacillus subtilis* ATCC 6633 as the test strain. Brain tissue was prepared for assay as previously described (3). The diluent for preparation of the brain homogenates was MH broth and this was added at a ratio of 1:1 (wt/vol). A standard curve was included on each plate, and, when serum levels exceeded the highest concentration of the standard, dilutions were made in normal rabbit serum until the zone of inhibition of the unknown fell within the limits of the curve. The zone of inhibition was plotted against the logarithm of the ampicillin concentration, and the results were used to generate a computer program for the calculation of unknowns by the method of least squares. Unknowns were assayed in duplicate, and zone sizes were measured in triplicate. The lowest concentration of ampicillin which could be accurately measured with this assay was 0.2 μg/ml (4).

TMP-SMZ assay. TMP, SMZ, and N^4-acetylated SMZ (AcSMZ) were assayed with high-pressure liquid chromatography. This methodology is detailed below. Brain tissue was processed as noted above.

AcSMZ (21) was kindly supplied by Hoffmann-La Roche, Inc. The metabolites of TMP (21) (N_1 and N_3 oxides of TMP, α-OH TMP, and 4-OH TMP) and the internal standard diaveridine were generously supplied by Burroughs-Wellcome Co. Glass-distilled, high-pressure liquid chromatography-grade methanol, chloroform, acetonitrile, and 2-propanol were purchased from Burdick & Jackson Labs, Muskegon, Mich. Mono- and dibasic sodium phosphate were ACS reagent grade and were obtained from Fisher Scientific Co. (Fairlawn, N.J.).

A high-pressure liquid chromatograph (model 204; Waters Associates, Inc., Milford, Mass.), equipped with a model U6K injection loop and an ultraviolet monitor (model 440) operated at 254 nm (SMZ-AcSMZ assays) or at 280 nm (TMP assay), was used. The reverse-phase column used was a prepacked stainless steel column (30 cm; 4-mm inner diameter) containing Bondapak-C_{18} 10 μ packing (Waters Associates, Inc.) generating 12,000 plates per meter. The mobile phase consisted of 25% methanol in pH 5.9 Sorensen phosphate buffer (0.05 M) which was filtered and degassed before use. The flow rate was set at 2.5 ml/min, and the high-pressure liquid chromatography attenuation was set at 0.01 absorbance units full scale (SMZ-AcSMZ assays) or at 0.005 absorbance units full scale (TMP assay).

Figure 1 shows the results of the high-pressure liquid chromatography assay for SMZ-AcSMZ in serum, CSF, and brain. The retention times for SMZ, AcSMZ, and diaveridine were 3.2, 3.8, and 7.0 min, respectively. Linear calibration plots of peak height ratio (SMZ or AcSMZ to diaveridine) versus concentration were constructed daily by subjecting standard biological samples containing 10 to 115 μg of SMZ and AcSMZ per ml to the assay methodology. TMP and its metabolites, at concentrations as high as 5 μg/ml, did not interfere with SMZ-AcSMZ determinations. In this study SMZ concentrations are equal to nonacetylated SMZ.

Figure 2 shows the results of the high-pressure liquid chromatography assay for TMP in serum, CSF, and brain homogenates. The retention times for TMP, the N_1 and N_3 oxides of TMP, α-OH TMP, 3-OH

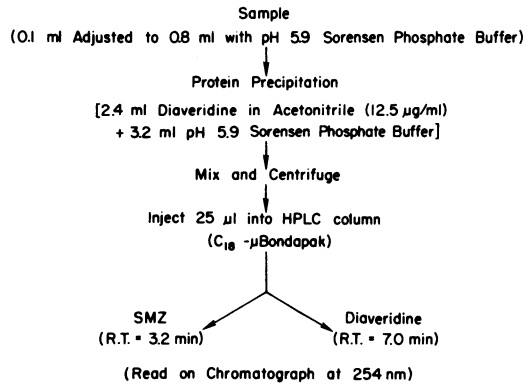

FIG. 1. *Outline of the methodology for determination of SMZ and AcSMZ concentrations in serum, CSF, and brain by high-pressure liquid chromatography.*

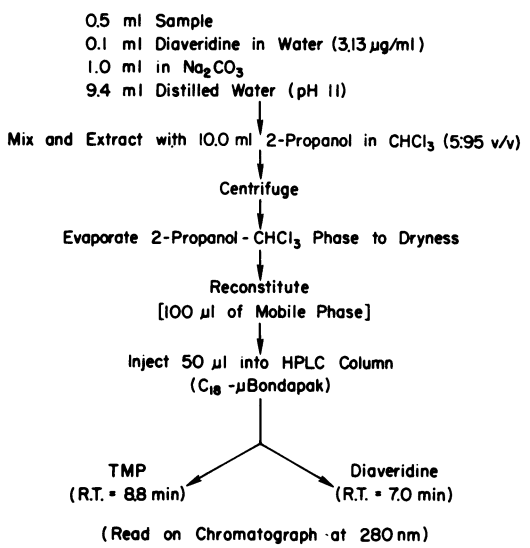

FIG. 2. *Outline of the methodology for determination of TMP concentration in serum, CSF, and brain by high-pressure liquid chromatography.*

TMP, 4-OH TMP, and diaveridine were 8.8, 6.6, 10.9, 4.1, 4.6, 3.4, and 7.0 min, respectively. Linear calibration plots of peak height ratio (TMP/diaveridine) versus concentration were constructed daily by subjecting standard biological samples containing 0.12 to 1.2 μg of TMP per ml to the assay methodology. SMZ and AcSMZ at concentrations as high as 115 μg/ml did not interfere with the determination of TMP.

The following assay modification was made for CSF samples containing low concentrations of TMP. One hundred microliters of a solution of diaveridine in acetonitrile (0.0625 μg/ml) was evaporated to dryness under nitrogen, and the residue was reconstituted with 250 μl of the CSF sample; 200 μl of the resultant mixture was injected onto the column.

The mean overall recoveries (n = 16 to 20 ± stan-

dard deviation) of SMZ, AcSMZ, and TMP were 102 ± 4.1, 98.5 ± 2.4, and 90.6 ± 2.1%, respectively.

CSF and brain tissue concentrations of TMP-SMZ and ampicillin were corrected for contamination with blood by measurement of hemoglobin concentration by the method of Lowry and Hastings (12) and a calculation utilizing hematocrit.

Percent CSF or brain penetration of a drug was calculated by the formula: (8-h CSF or brain concentration/8-h serum concentration) × 100.

Statistical analysis. Differences between control and treatment groups were assessed by Student's *t* test. A *P* value of <0.05 was considered significant.

RESULTS

In vitro studies. Both strains of *E. coli* were susceptible to ampicillin and TMP-SMZ by the Kirby-Bauer technique. MICs and MBCs of ampicillin and TMP-SMZ for the test strains are given in Table 1.

Course of infection in untreated rabbits with EC7 meningitis. Nine rabbits were inoculated intracisternally with EC7, not treated, and followed to determine duration of survival. Eight rabbits survived 18 h or longer: two died at 18 h, one died at 22 h, and the remaining five died 50 to 88 h post-inoculation (mean survival, 55 h). At 16 h after inoculation the bacterial count of EC7 in the CSF of these eight rabbits ranged from 3.0×10^4 to 10^7 CFU/ml (mean, 8.5×10^5 CFU/ml). One rabbit died 8 h after inoculation and had 4×10^8 CFU of EC7 per ml of CSF at the time of death. This early death can possibly be attributed to overwhelming infection due to inadvertent inoculation with an excessive number of bacteria. Thus, EC7 meningitis was fatal for nine untreated rabbits within 96 h of inoculation. Animals inoculated with EC7 were clinically ill as manifested by rhinnorhea, conjunctivitis, nuchal rigidity or opisthotonus, and hyperventilation.

Antimicrobial levels. Table 2 shows a comparison of TMP and SMZ levels in serum, CSF, and brain in rabbits with and without meningitis at the end of 8 h of treatment. There was no statistical difference in mean serum, CSF, and brain concentrations of TMP and SMZ between rabbits with EC7 and EC13 meningitis. Therefore, these data were pooled and used to calculate the mean concentrations reported in Table 2.

Mean serum TMP and SMZ concentrations in rabbits with meningitis were similar to those in controls. The mean concentration of TMP in CSF of rabbits with meningitis was greater than that found in the two control rabbits. For most of the rabbits with meningitis there was a twofold or greater increase in CSF TMP levels compared with those in controls. The concentration of SMZ in CSF of rabbits with meningitis showed considerable variability, but the mean level was similar to that of the two controls. Of note are CSF and brain TMP and SMZ concentrations in the two control rabbits that exceeded the MBCs of these drugs for the test strains.

Ampicillin concentrations in serum, CSF, and brain of rabbits with meningitis are also pre-

TABLE 1. *MICs and MBCs of ampicillin and TMP-SMZ for EC7 and EC13*

Strain	MIC (μg/ml)		MBC (μg/ml)	
	Ampicillin	TMP-SMZ	Ampicillin	TMP-SMZ
EC7	1.56	0.02-0.4	3.13	0.04-0.78
EC13	0.5	0.04-0.78	2.0	0.078-1.56

TABLE 2. *Antimicrobial concentrations in serum, CSF, and brain of rabbits with and without meningitis after 8 h of therapy*

Determination	Rabbits without meningitis[a]		Rabbits with EC7 and EC13 meningitis[b]		
	TMP	SMZ	TMP	SMZ	Ampicillin
Serum (μg/ml)	4.50, 8.98	65.1, 66.8	4.66 ± 2.46 (18)[c]	60.7 ± 34.0 (18)	37.3 ± 15.4 (17)
CSF (μg/ml)	0.17, 0.44	15.1, 13.6	0.80 ± 0.41 (17)[d]	15.7 ± 12.1 (18)	2.6 ± 1.7 (15)[e]
Brain (μg/ml)	0.12, 0.20	3.0, 2.2	0.23 ± 0.7 (10)[f]	3.31 ± 3.3 (10)	0.6 ± 4 (8)[g]
Mean % CSF penetration	3.8, 4.8	23.2, 20.4	18.0 ± 7.8 (17)	27.2 ± 12.4 (18)	8.9 ± 6.4 (15)
Mean % brain penetration	2.6, 2.3	4.6, 3.2	6.6 ± 3.8 (10)	5.5 ± 3.4 (10)	1.9 ± 1.5 (8)

[a] Results of levels measured in two rabbits; ampicillin was not given to rabbits without meningitis.
[b] Results expressed as mean ± 1 standard deviation.
[c] Numbers in parentheses indicate the number of rabbits from which specimens were available for analysis.
[d] In one rabbit there was an insufficient amount of CSF to perform TMP assay.
[e] In two rabbits there was insufficient CSF to perform the assay.
[f] Brain levels measured in 10 animals with EC7 meningitis only.
[g] Brains were removed and ampicillin levels were performed in 8 of 17 rabbits.

sented in Table 2. For both models of meningitis, mean ampicillin levels in the serum and CSF were similar, and the data were again pooled. The mean ampicillin concentration in CSF exceeded the MIC for both test strains. The mean ampicillin concentration in brain homogenate of eight animals exceeded the MIC for EC13 but not EC7.

Therapy of experimental meningitis. Table 3 shows the effect of an 8-h infusion of TMP-SMZ and ampicillin on bacterial counts in CSF compared with controls given saline. The mean number of organisms in CSF 16 h after inoculation was the same for each of the three groups for both test strains (range, 10^3 to 4.7×10^8 CFU/ml, $n = 52$).

In rabbits with EC7 meningitis, TMP-SMZ and ampicillin therapy resulted in a significant decrease in the mean concentration of bacteria in CSF when compared with that of controls. The mean decrease in bacterial counts was the same for rabbits treated with TMP-SMZ and ampicillin. In rabbits with EC13 meningitis, TMP-SMZ and ampicillin therapy also resulted in a significant reduction in the mean number of bacteria in CSF compared with that of controls. The difference between these agents in the mean fall of bacterial counts after 8 h of therapy was not statistically significant.

During the 8-h period between 16 and 24 h post-inoculation, control rabbits with EC7 meningitis had a greater mean increase in bacterial counts than did rabbits with EC13 meningitis (2.5 logs versus 0.03 log). This difference occurred despite the fact that the mean bacterial concentration of EC7 in CSF 16 h after inoculation was less than that for EC13.

DISCUSSION

Penetration of TMP-SMZ into CSF and brain. In the absence of meningeal inflammation, ampicillin, cephalosporins, and aminoglycosides penetrate poorly, if at all, into CSF and the brain (3). In the presence of inflamed meninges, penetration of these agents into CSF increases and is proportional to the severity of infection (4), but may not exceed the MIC for a given pathogen.

In the current study TMP and SMZ concentrations in CSF exceeded the MBCs of both the test strains in the absence of an inflammatory stimulus. CSF concentrations further increased, especially that of TMP, with meningeal inflammation and were similar to those reported in humans (10, 26). This degree of penetration into the central nervous system by TMP-SMZ, coupled with its antimicrobial activity against aerobic gram-negative bacilli, may provide an advantage in treatment of bacterial meningitis caused by susceptible organisms.

At the doses used in this study, TMP-SMZ infusion produced measurable levels in brain tissue. This may have important implications for therapy of brain abscesses. A few case reports document successful treatment of brain abscesses with TMP-SMZ (8, 13). On the other hand, Neu (17) in a recent review of antimicrobial therapy of brain abscesses states that TMP-SMZ penetrates poorly into brain substance. The present study offers supporting evidence for the use of TMP-SMZ in therapy of brain abscesses since penetration into brain substance occurred without inflammation and was enhanced in the presence of an infectious process.

TABLE 3. *Effects of 8 h of antimicrobial therapy on bacterial counts in CSF of rabbits with EC7 and EC13 meningitis*

Infecting strain	Antibiotic	No. of animals	Bacterial concn (log_{10} CFU/ml) 16 h after inoculation	ΔMean log_{10} CFU/ml after 8 h of therapy[a]	
EC7	None	7	4.3 ± 1.0[b]	+2.5 ± 4.4	
					$P < 0.001$
	TMP-SMZ	10	5.2 ± 1.5	−5.2 ± 1.6	$P < 0.001$
					$P > 0.1$
	Ampicillin	9	5.1 ± 1.2	−5.1 ± 1.1	
EC13	None	9	5.1 ± 1.4	+0.03 ± 6.0	
					$P < 0.01$
	TMP-SMZ	8	5.7 ± 0.6	−5.7 ± 0.7	$P < 0.01$
					$P > 0.1$
	Ampicillin	7	4.8 ± 1.0	−4.8 ± 1.0	

[a] Log_{10} (CFU per ml of CSF 16 h after inoculation − CFU/ml after 8 h of therapy) ± 1 standard deviation; +, net increase; −, net decrease in the number of bacteria.

[b] Mean log_{10} CFU/ml ± 1 standard deviation.

Whether TMP-SMZ can penetrate into a cerebral abscess and maintain its antibacterial activity in this milieu requires further study.

Penetration of ampicillin into CSF and the brain. There was considerable variability in the ampicillin serum levels as demonstrated by the large standard deviation in Table 2. Other authors (20, 24, 25) have also noted wide animal-to-animal variability of serum levels when antibiotics are administered on the basis of weight. But an individual animal will have minimal fluctuations in the serum level with a constant infusion of drug (3). Therefore, to minimize variability one can present the data as compartmental ratios (CSF/serum, brain/serum), which has been done in Table 2.

In a previous study (3) it was shown that there is a 9- to 10-fold increase in the concentration of ampicillin in CSF of rabbits with EC13 meningitis compared to those without meningitis. This resulted in a mean percent penetration into the CSF of 9.7% of the simultaneous serum level. In the present study, mean percent penetration of ampicillin into CSF of rabbits with meningitis was 8.9%. This produced concentrations of ampicillin in CSF which exceeded the MIC for both test strains and correlated with a marked reduction in CSF bacterial counts of EC7 and EC13.

There is minimal information about the penetration of ampicillin or other antibiotics into brain tissue. Kramer et al. (11) have reported on the penetration of several antibiotics, including ampicillin, into normal brain tissue of patients undergoing neurosurgical procedures. In this study, the mean percent penetration of ampicillin into brain tissue was 1.9% after intravenous administration of 2 g of ampicillin. Among four of five patients sampled less than 2 h after the ampicillin injection, no drug could be detected in brain samples.

In the present study the mean percent penetration of ampicillin into brain tissue of rabbits with meningitis was 1.9% at the completion of an 8-h continuous infusion. One cannot directly compare the results of these studies because of differences in dose, method of administration, half-life, and methods for determining ampicillin concentration. Data in the present study reveal minimal penetration of ampicillin into brain tissue of rabbits in the presence of meningitis. Whether the concentration of ampicillin would be increased further in the presence of parenchymal brain infection and the drug would maintain its antibacterial activity requires further study.

Therapy of meningitis. In rabbits with EC7 and EC13 meningitis, therapy with TMP-SMZ for 8 h reduced the concentration of bacteria in CSF the same extent as ampicillin therapy for 8 h. This was expected based on the in vitro data and the degree of penetration of these agents into the CSF of rabbits with meningitis. Since all animals were sacrificed after 8 h of treatment, the possibility that either of these regimens would result in cure of the meningitis could not be determined in this study. Nonetheless, the rapid reduction in bacterial counts in CSF demonstrated in this study after only 8 h of therapy may be important for the successful therapy of bacterial meningitis (14).

During the 8-h period between 16 and 24 h post-inoculation, the mean bacterial concentration in the CSF of animals given saline increased by 2.5 logs for EC7 but by only 0.03 log for EC13. The reason for this difference in growth during this period is not clear. The mean concentration of EC13 in CSF was greater than that of EC7 at 16 h post-inoculation but this difference was not statistically significant. The initial inoculum for both strains was also the same. One possible explanation is that the EC7 strain, unlike the EC13 strain, contains the K1 antigen. This antigen has been said to confer increased virulence (28). However, K1 antigen had no implications for survival since the mean duration of survival for untreated rabbits with EC7 and EC13 meningitis was similar (55 h versus 61 h).

These results can be extrapolated to the clinical setting only with great caution. First, a large inoculum is required to produce a lethal infection in this model of meningitis. Second, the route of infection and pathogenesis differ from meningitis which occurs in humans. Third, the impact of preexisting host defenses has not been analyzed in these studies.

In conclusion, TMP and SMZ given parenterally penetrated well into CSF and the brain in the presence or absence of meningeal inflammation in rabbits. In two models of ampicillin-susceptible *E. coli* meningitis, continuous intravenous TMP-SMZ therapy produced bacterial killing in CSF equivalent to that of ampicillin. These findings suggest a potential therapeutic role for parenteral TMP-SMZ in the treatment of *E. coli* meningitis and possibly for meningitis caused by other susceptible bacteria, including ampicillin-resistant organisms.

ACKNOWLEDGMENTS

This study was supported by the Veterans Administration and a grant from Hoffmann-La Roche, Inc.

We thank J. Anderson for her technical assistance and S. L. Maier for her secretarial work.

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