THE JOURNAL OF INFECTIOUS DISEASES • VOL. 149, NO. 4 • APRIL 1984
 0022-1899/84/4904-0015$01.00

The Postantibiotic Effect in the Treatment of Experimental Meningitis Caused by *Streptococcus pneumoniae* in Rabbits

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The relevance of a postantibiotic effect in the treatment of pneumococcal meningitis was evaluated in a rabbit model. After administration of a single intravenous bolus of ampicillin at various dosages, such an effect was observed in all animals. The duration of this effect in vivo (2.5–18 hr) was consistently longer than that in vitro (1–4.3 hr); however, in rabbits the postantibiotic effect was eliminated by the administration of intravenous plus intracisternal β-lactamase. In an assessment of the potential therapeutic benefit of the postantibiotic effect, the efficacy of two regimens of treatment with different intervals between doses was compared. One group of animals received ampicillin every 4 hr and another every 12 hr. With sufficiently high doses, drug concentrations in cerebrospinal fluid exceeded the minimal bactericidal concentration for most of the 4-hr interval but for only about one-third of the 12-hr interval. The rate of cure was similar for the two regimens and approximated 100% when peak drug concentrations in cerebrospinal fluid exceeded the minimal bactericidal concentration by at least 10-fold.

The factors that influence efficacy of the treatment of infectious diseases with antibiotics are not well defined [1]. Complex interactions between host defense mechanisms, bacteria, and antibiotics and difficulties associated with the accurate measurement of drug concentrations at the site of infection make it almost impossible to determine the exact relation between treatment and effect. Hence, the relative importance of such pharmacokinetic variables as peak drug concentration, total dose, period during which drug concentrations exceed the MIC, and interval between doses has not been studied extensively with regard to most infections.

Bacterial meningitis has several distinguishing features that make its treatment particularly complex. The blood-CSF barrier limits the penetration of antibiotics into the CSF [2]; active transport systems rapidly clear most β-lactam drugs from the CNS [3, 4]; and impaired host-defense mechanisms in the CSF make bactericidal activity important [5, 6]. Since bacterial meningitis is such a devastating disease, the regimens currently recommended consist of large doses of antibiotics given either at frequent intervals or by constant infusion. However, with respect to the persistently high morbidity and mortality associated with the disease [7, 8] and the potential toxicity of the antibiotics used to treat it, a more detailed understanding of the principles operative in therapy for meningitis seems important.

We have shown [9] that two intermittent doses of penicillin (with an interval of 4 hr) are as effective as a constant infusion of the same amount of drug over 8 hr in reducing bacterial counts in rabbits with pneumococcal meningitis. The fact that trough drug levels below the MBC for the infecting organisms in 64% of CSF samples did not cause any apparent loss of bactericidal activity (compared with the activity of the drug at levels that were constantly above the MBC) suggested that a postantibiotic effect (PAE) exists in CSF in vivo. The occurrence of a PAE, defined as delayed regrowth of bacteria after limited exposure to antibiotics, was recognized early in the antibiotic era [10–12]. Eagle and colleagues documented this phenomenon in vivo and evaluated its potential role in the treatment of bacterial infections [13].

Received for publication [editor will add date in galleys].

We thank Sämi Kunz and Julian Vaxelaire for technical assistance and Rebecca Brooks-Fournier for help with preparation of the manuscript.

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However, the relevance of the PAE for the treatment of meningitis has not been established.

We designed the present study to characterize the PAE in an animal model of pneumococcal meningitis and to determine its potential implications for therapy. We examined the relation between drug concentration in the CSF and bacterial growth kinetics at this site over time after a single iv injection of ampicillin. To evaluate the possibility that the observed PAE was due to undetectable drug in the CSF, we characterized the bacterial growth curves in animals receiving a β-lactamase during the PAE. In order to determine the possible impact of the PAE on therapy, we examined the relative influence of several pharmacokinetic variables (dose, peak CSF concentration, interval between doses) on the outcome of treatment.

Materials and Methods

Organism. A strain of *Streptococcus pneumoniae* type 3 originally isolated from a human adult with meningitis and characterized earlier [14] was used in all experiments. The strain was grown in trypticase soy broth (TSB; Difco Laboratories, Detroit) supplemented with 50% inactivated calf serum in a moist atmosphere of 5% CO_2–95% air at 37 C for 18 hr. The culture was then centrifuged for 10 min at 7,000 *g*, resuspended in 0.9% NaCl, and stored in 1-ml portions containing $\sim 10^7$ cfu in the vapor phase of a liquid-nitrogen vessel.

Viable counts. Viable bacterial counts were determined in trypticase soy agar (TSA; Difco) supplemented with either 5% fresh sheep blood or 5% human blood. Plates were examined after incubation at 37 C in a moist atmosphere of 5% CO_2–95% air for 18–24 hr. The limit of detectability was 10 cfu/ml for 0.1 ml of the culture in broth (in vitro tests) or CSF.

Antibiotics. A stock solution of 10 mg of ampicillin/ml (Penbritin®; Beecham, Bristol, Tenn) in 0.9% NaCl was prepared daily and further diluted in 0.9% NaCl if required.

Inactivation of ampicillin. Penicillin-amido-β-lactamhydrolase (E.C. 3.5.2.6.; Whatman Biochemicals, Maidenhead, Berkshire, England) was used for hydrolysis of ampicillin in the in vitro experiments. The final concentration of β-lactamase was 0.8 unit/ml of agar or 1.6 units/ml of culture broth [15, 16]. In rabbits, ampicillin was inactivated by iv and intracisternal injection of penicillin-amido-β-lactamhydrolase (Riker Laboratories, Northridge, Calif); each animal simultaneously received 300,000 IU by the iv route and 100,000 IU intracisternally. The activity of β-lactamase in CSF was confirmed by the demonstration that the CSF of control animals given β-lactamase, as already described, completely inactivated ampicillin in vitro.

Antibiotic assay. Concentrations of ampicillin were determined by an agar well-diffusion technique [17], with *Micrococcus luteus* ATCC 9341 (*Sarcina lutea*) as the test organism. Plates (diameter, 85 mm) were filled with 15 ml of antibiotic medium no. 1 (Oxoid, London) containing 10^7 viable cells/ml of agar. Wells (diameter, 8 mm) were cut and filled with 0.05 ml of CSF sample or standard dilutions, and zones of inhibition were measured after incubation overnight at 37 C. CSF standards were prepared in 0.9% NaCl, which has been shown to produce zones identical to those in infected or uninfected CSF [9].

In vitro studies. The MICs and MBCs of ampicillin were determined by a microtiter method in heart infusion broth with bacterial inocula of 10^4 and 10^6 cfu [14, 18]. The MIC was defined as the lowest drug concentration that inhibited visible bacterial growth and the MBC as the lowest concentration that killed 99.9% of the organisms over 24 hr. MICs and MBCs were also determined in pooled rabbit CSF and in broth with a pH of 6.8.

The effects of in vitro exposure of the organisms to ampicillin were examined by a time-kill regrowth method [19]. All experiments were performed in TSB plus 50% calf serum by shaking at 200 cycles/min (Mini-Shaker®; A. Kuhner, Basel, Switzerland). A stock culture of *S pneumoniae* type 3 was grown for 4 hr in order to obtain organisms in the logarithmic phase of growth. Portions (10 ml) of this culture were transferred into 25-ml flasks containing either ampicillin solutions in 0.9% NaCl or pure 0.9% NaCl (control flasks). The time of the transfer was defined as time zero. Ampicillin was removed at intervals by centrifugation three times for 10 min at 1,500 *g* [20] or by inactivation with β-lactamase. Fresh, prewarmed TSB supplemented with 50% serum was used for resuspension of bacteria after centrifugation. Viable cell counts were performed on the original inoculum before exposure, at the end of the period of exposure to the drug, and thereafter at required intervals during the regrowth phase. The PAE was

calculated as the time necessary for the cultured inoculum to increase by 1 $\log_{10}$ cfu/ml after removal of the antibiotic minus the time required by a control, unexposed culture to increase by 1 $\log_{10}$ cfu/ml. The same experiment was performed with pooled CSF (instead of supplemented TSB) and β-lactamase for inactivation of ampicillin.

Rabbit model of meningitis. New Zealand white and Chinchilla rabbits weighing 2–3 kg were used for the induction of pneumococcal meningitis, as described previously [21]. The rabbits were anesthetized with pentobarbital (30 mg/kg), and a dental acrylic helmet was attached to their skulls by four screws. At least 24 hr later, the animals were again anesthetized and attached by the helmet to a stereotactic frame. A Quincke spinal needle (25 gauge, 3½ inches (Becton, Dickinson, Rutherford, NJ) was introduced atraumatically into the cisterna magna with a geared electrode inducer, and the animals were infected with an inoculum of ~2 × 10^5 cfu of *S pneumoniae* in 0.2 ml. The needle was removed, and the animals were returned to their cages. Eighteen hours later all animals demonstrated signs of meningitis (temperature of ≥39.6 C, lethargy, CSF pleocytosis, and positive CSF culture results) [6, 9, 21].

PAE in vivo. At 18 hr the animals were anesthetized with 1.5 g of iv-administered urethane/kg (Merck Sharp and Dohme, Rahway, NJ). The spinal needle was again introduced into the cisterna magna, and CSF was withdrawn serially every 2–3 hr for 24 hr for determination of bacterial titers and drug concentration in CSF. At time zero the animals received an iv injection of ampicillin at one of the following doses (in mg/kg): 2, 3, 4, 6, 12.5, 20, or 62.5. In six animals the ampicillin was inactivated by the iv injection of β-lactamase (300,000 IU by the iv route and 100,000 IU by the intracisternal route) 2 or 4.5 hr after drug administration.

Treatment with different regimens. Animals were prepared as already described, and CSF for cultures was obtained by spinal taps performed 18 hr after infection. Animals were then assigned to one of 10 treatment groups or to a control group that received no treatment. Treatment consisted of either two iv injections of ampicillin 12 hr apart or four iv injections every 4 hr; thus, both regimens were completed after 12 hr. The dose in each injection was one of the following (in mg/kg): 4.17, 6.25, 12.5, 25, or 37.5. Peak levels of drug in CSF were determined 30 min after the first injection. Animals were observed every 4 hr during the experiment (with temperatures determined) and underwent final evaluation (including sampling of CSF for culture) 36 hr after the last dose. After each spinal tap the needle was removed.

Results

Susceptibility. The MIC and MBC of ampicillin for the test strain were 0.06–0.08 μg/ml with an inoculum of 10^4 cfu/ml and 0.1–0.125 μg/ml with an inoculum of 10^6 cfu/ml. MICs and MBCs were identical in broth at pH 7.4, in broth at pH 6.8, and in CSF ex vivo.

Characteristics of the PAE in vitro. Figure 1 shows the principal characteristics of growth of *S pneumoniae* in the presence and absence of ampicillin. In the absence of antibiotics, the bacteria grew promptly (in 4 hr) from an initial count of 6.2 $\log_{10}$ cfu/ml to 8 $\log_{10}$ cfu/ml. The presence of ampicillin at concentrations at or above the MBC led to a uniform decline in bacterial titers of 1.5 $\log_{10}$ cfu/ml during the first 2 hr. The rate of killing was constant over a wide range of ampicillin concentrations (1–100 times the MBC). Removal of the antibiotic after exposure of the organisms for 2 hr resulted in delayed regrowth (i.e., a PAE) in all experiments. Whether ampicillin was inactivated by β-lactamase or removed by centrifugation and washing, the duration of the PAE in vitro was the same. Higher drug levels resulted in a longer PAE (figure 1). The lowest drug concentration that resulted in a measurable PAE was 0.03 μg of ampicillin/ml (one-half the MIC), with a PAE duration of 1 hr. At a concentration of 10 μg of ampicillin/ml, the PAE lasted for 4.3 hr. When the same experiments were performed in pooled rabbit CSF, the level of initial killing during the 2-hr period of ampicillin exposure was markedly reduced (i.e., to 0.5 $\log_{10}$ cfu/ml), and the average generation (or doubling) time of bacterial growth was increased (from 21 min in broth to 37 min in CSF). The duration of the PAE, however, was the same in broth and CSF.

Characteristics of the PAE in vivo. The dynamic relation between drug concentrations and bacterial titers in CSF was examined in 17 animals that had been infected intracisternally 18–20 hr prior to the iv administration of ampicillin. The administration of different drug doses (1–62.5

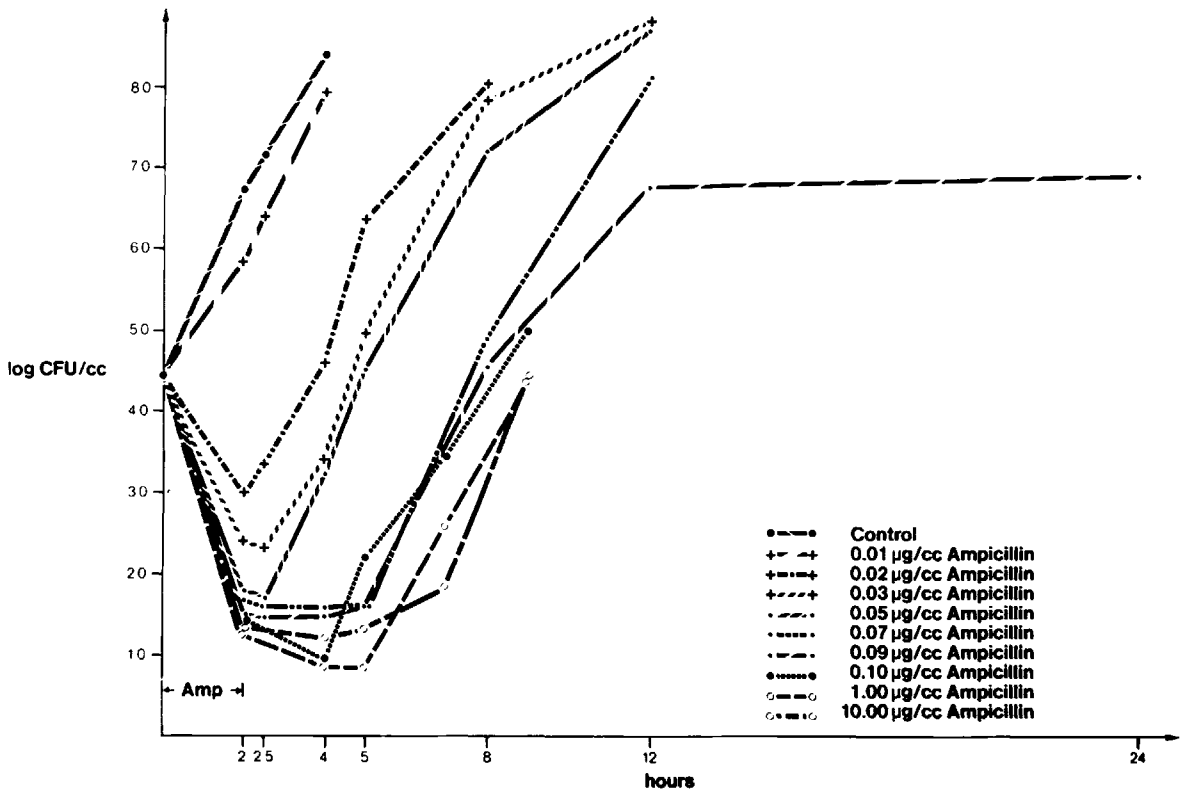

Figure 1. Effect of exposure for 2 hr to different concentrations of ampicillin on growth curves of *S pneumoniae* in broth. Bacterial titers are plotted as $\log_{10}$ cfu/ml against time (hr). The control culture contained no ampicillin. After exposure for 2 hr to the indicated concentrations of ampicillin, the drug was removed from the culture by centrifugation and three washes.

mg/kg) resulted in a wide range of CSF concentrations (0.04–6.4 µg/ml); the dose administered and the resulting peak CSF concentrations correlated well (r = .921; P < .001). The dose and the peak CSF concentrations also correlated (r = .616; P < .02) with the period during which CSF concentrations were above the MIC (median, 4 hr; range, 1–7.5 hr).

The mean bacterial titer (± SD) in CSF was 6.0 (± 1.0) $\log_{10}$ cfu/ml at the time of drug administration. The injection of ampicillin was followed by a median drop in bacterial titers of 4.4 $\log_{10}$ cfu/ml (range, 1.9–6.5 $\log_{10}$ cfu/ml). This decline in bacterial titers correlated with the dose of ampicillin administered (r = .619; P < .01).

The total drug effect, defined as the period from the injection of ampicillin to the beginning of bacterial regrowth (figure 2), lasted for a median of 11 hr (range, 3.5–20 hr) and correlated with the ampicillin dose (r_s = .488; P < .05). In animals receiving doses of ampicillin that produced peak CSF concentrations of ≤10 times the MBC, the total duration of the drug's effect consistently exceeded 12 hr. The PAE, defined as the total duration of the drug's effect minus the period during which drug concentrations were above the MIC (figure 2), ranged from 2.5 hr to 18 hr (median, 6.5 hr) and did not correlate with either the dose of ampicillin or the peak concentration of ampicillin in CSF (r = .050; P = not significant).

Effect of β-lactamase on the PAE in vivo. In six additional animals that were prepared identically, the administration of an iv bolus of 12.5 mg of ampicillin/kg was followed by the injection of 400,000 IU of β-lactamase (300,000 IU by the iv route and 100,000 IU into the cisterna magna) after 2 hr or 4.5 hr. In all animals the injection of β-lactamase reversed the PAE. In both treatment groups, bacterial titers in CSF had increased by an average of 0.5 $\log_{10}$ cfu/ml 0.5 hr after administration of the enzyme and by 0.9 $\log_{10}$ cfu/ml 1 hr after the injection (figure 3).

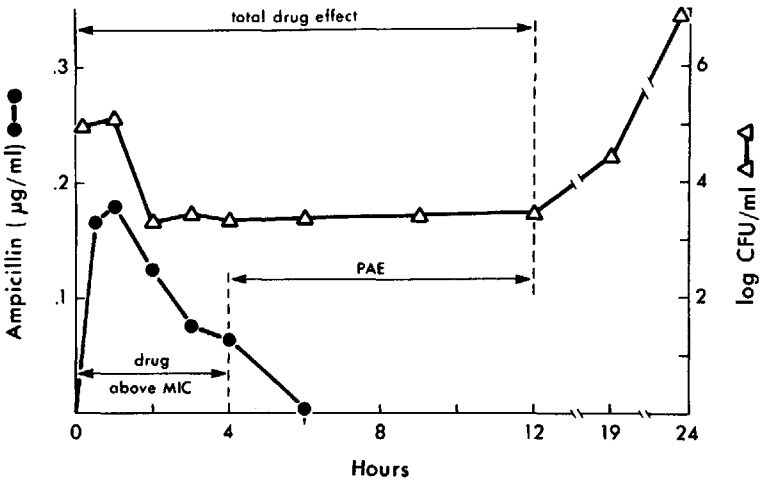

Figure 2. Relation of ampicillin concentrations and bacterial titers in CSF of a representative rabbit that received 12.5 mg of ampicillin/kg iv at time zero. Drug concentrations and bacterial titers were determined in serial samples of CSF.

Influence of different regimens on therapeutic efficacy. A total of 235 rabbits with meningitis were used in studies of the influence of different doses of ampicillin and different intervals between doses on sterilization of CSF cultures 36 hr after the last injection. The 28 untreated control rabbits all died within 66 hr of bacterial inoculation or had positive CSF culture results (mean, 10^5 cfu/ml) and fever (≥39.6 C); no spontaneous cures were observed. The remaining 207 animals were assigned to the various treatment groups. Therapy was initiated 18 hr after infection, when 100% of the animals had fever and positive CSF culture results (3.9 ± 1.2 $\log_{10}$ cfu/ml). Forty-nine rabbits had to be excluded from the evaluation either because they died before the completion of the 66-hr experiment (n = 15) or because no CSF could be obtained for the final evaluation (n = 34). The majority of the early deaths resulted from experimental procedures. Since evaluation of the results depended on the result of culture of the final CSF sample, data on all animals that could not be classified with certainty for one of the reasons mentioned were excluded. Even if all deaths had been counted as treatment failures, the outcome would not have been significantly changed.

The two most important factors determining outcome in our experiments were the dose in a single injection of ampicillin and the peak drug concentrations in CSF. As previously stated, these two variables are closely related. Small single doses, which produced peak CSF concentrations of 0.2–0.5 μg/ml (two to five times the MBC), inconsistently resulted in sterile CSF; in contrast, large doses, with peak CSF concentrations of >10 times the MBC, sterilized the CSF of >90% of the animals, regardless of other variables in the treatment regimen (table 1). When regimens consisting of the same single dose but different total doses and intervals between doses (four doses at 4-hr intervals vs. two doses at 12-hr intervals) were compared, the more frequent dosing regimen appeared to be preferable but differences did not reach statistical significance ($P \geq .2$). On the other hand,

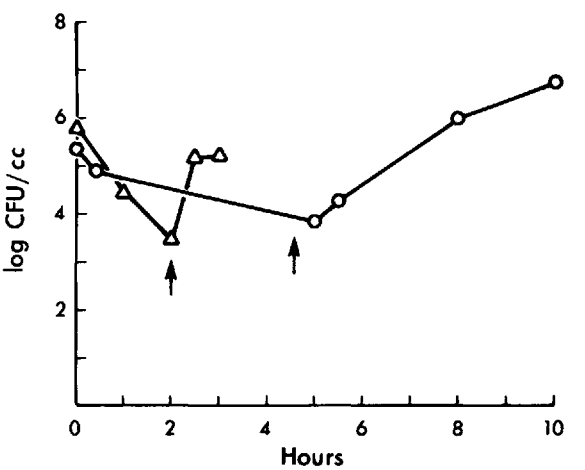

Figure 3. Effect of inactivation of ampicillin in CSF by injection of β-lactamase (↑) in two representative rabbits. Each animal received 12.5 mg of ampicillin/kg iv 18 hr after being infected. Bacterial titers of *S pneumoniae* were determined in CSF over time. The animals received 400,000 IU of β-lactamase (300,000 IU by the iv route and 100,000 IU intracisternally) 2 hr and 4.5 hr after the injection of ampicillin.

Table 1. Results of ampicillin treatment of meningitis caused by *S pneumoniae* in rabbits.

Single dose (mg/kg)	No. of doses	Total dose (mg/kg)	Mean peak drug level in CSF (± SD)	No. of animals with sterile CSF/ no. evaluated (% with sterile CSF)*
0	0	0	0	0/28 (. . .)
4.17	2	8.34	0.20 ± 0.09	0/10 (. . .)
4.17	4	16.68	0.20 ± 0.14	1/9 (11)
6.25	2	12.5	0.20 ± 0.08	4/19 (21)
6.25	4	25	0.28 ± 0.16	9/21 (43)
12.5	2	25	0.52 ± 0.37	13/23 (57)
12.5	4	50	0.54 ± 0.35	19/24 (79)
25.0	2	50	1.14 ± 0.54	21/22 (95)
25.0	4	100	1.44 ± 0.46	8/8 (100)
37.5	2	75	1.44 ± 1.26	11/12 (92)
37.5	4	150	1.55 ± 0.89	10/10 (100)

* Each pair of results listed consecutively in the table was compared by the χ^2 test or the Fisher's exact test. In all cases the differences between values were insignificant ($P > .05$).

when regimens with the same total dose were compared, those involving drug administration every 12 hr (which resulted in higher individual peak CSF concentrations) were found to be slightly advantageous but not significantly so ($P \geq .2$).

Another variable that may influence the bacteriologic response is the proportion of the interval between doses during which CSF concentrations exceed the MIC. Our preliminary experiments showed that CSF concentrations are above the MIC for ~4 hr after an injection of 12.5 mg of ampicillin/kg and for ~6 hr after a dose of 37.5 mg/kg. Thus, regimens with single doses of 12.5 mg/kg administered every 4 hr result in CSF concentrations above the MIC for 100% of the interval between doses (i.e., the whole period of treatment), while those with single doses of 37.5 mg/kg administered every 12 hr produce CSF concentrations that exceed the MIC for (at most) 50% of the time. Our results did not show a significant influence of the interval between doses on the results of treatment. However, when the single dose administered every 12 hr was increased from 12.5 mg/kg to 25 mg/kg, the outcome of treatment was significantly better ($P < .01$), even though the period during which CSF concentrations were above the MIC increased only from 30% to 40% of the interval between doses. Thus, our results indicate that the period during which CSF concentrations exceed the MIC is not a major determinant of outcome within the limits examined (i.e., with CSF concentrations above the MIC for >30% of the interval between doses).

Thus, the size of the single dose of ampicillin and the peak concentration of the drug in CSF determine the bacteriologic result in our animal model of meningitis. Doses that result in peak CSF concentrations of >10 times the MBC are highly effective, with sterile CSF in >90% of cases. The total amount of drug given during the period of treatment, the interval between doses, and the period during which the drug concentration in the CSF is above the MIC appear to be of minor importance.

Discussion

Early in the antibiotic era, Parker and colleagues [10, 11] observed that the inhibitory effect of penicillin on staphylococci in vitro persisted for some time after removal of the drug. This PAE has since been reproduced in vitro with many different organisms and antibiotics [12, 19, 22]. A few years after Parker's initial observations, Eagle and colleagues conducted a series of experiments on the importance of several variables of antibiotic treatment, including the PAE, in mice and rabbits [13, 23, 24]. These investigators found that a PAE similar to that found in vitro also existed in vivo, that an increase of doses above a critical value did not enhance the bactericidal action of penicillin, and that the most important variable determining outcome was the period during which the concentration of drug was maintained above the MIC. Interest in these questions has persisted. Most recently, studies conducted with a thigh model of pneumococcal infection in neutropenic mice demonstrated that penicillin produced a PAE only after the ad-

ministration of large doses and prolonged exposure, whereas erythromycin and tetracycline produced a PAE similar to that observed in vitro [25]. Furthermore, the period during which drug concentrations were above the MIC was found to be the most important factor determining the results of treatment in these experiments [25, 26]; these observations were similar to those of Eagle et al. [23, 24]. In another study Bakker-Woudenberg and associates examined the effectiveness of penicillin against pneumococcal pneumonia in decomplemented rats and confirmed the importance of prolonged periods during which drug concentrations exceeded the MIC for maximal therapeutic benefit [27].

The understanding of pharmacokinetic factors that influence the efficacy of therapy for bacterial meningitis is incomplete. Preliminary studies in a rabbit model of pneumococcal meningitis have suggested that a PAE exists in CSF [9], but the role of this PAE and its relation to other variables, such as dose and duration of drug exposure, have not been investigated.

The PAE documented in our studies in vivo of CSF differed in several respects from the PAE observed in vitro. The former lasted considerably longer than the latter (2.5–18 hr vs. 1–4.3 hr) and was considerably more variable. The duration of the in vivo PAE did not correlate with the dose or the peak concentration of ampicillin in CSF and could be reversed by the administration of β-lactamase, which inactivated residual drug in the subarachnoid space.

These characteristics suggest that the PAE observed in our animal model of meningitis may be a different phenomenon from the PAE observed in vitro. The strongest evidence for this hypothesis derives from the striking effect of β-lactamase administration on the PAE in vivo. Our findings indicate that the in vivo effect is the consequence of the presence of small residual amounts of drug in CSF. When this drug is inactivated, no further PAE can be documented. This result is in contrast to that in vitro, where the PAE was found (in both this study and that of McDonald et al. [19]) to be unaffected by β-lactamase.

If the PAE in CSF is indeed attributable to the presence of small residual amounts of drug, its duration should reflect the pharmacokinetic characteristics of drug elimination from CSF. In earlier studies we found considerable variation of CSF pharmacokinetics in infected rabbits [14, 28], and the slow gamma phase of drug elimination is probably subject to similar variability. Furthermore, the duration of the gamma phase is probably a function not only of the initial CSF concentrations but also of other factors that can influence CSF pharmacokinetics (e.g., degree of inflammation, rate of CSF turnover). Thus, the observed variability of the PAE and its lack of correlation with the initial CSF peak in vivo may simply be the expressions of pharmacokinetic differences that affect the duration of the gamma phase of drug elimination in individual rabbits.

In the absence of active drug in CSF (i.e., after β-lactamase administration), we did not find a PAE in our model. This result is in agreement with the findings of other researchers who, examining the occurrence of a PAE in neutropenic mice infected with *S pneumoniae*, found either no PAE [26] or only a short PAE after prolonged exposure to large doses of penicillin [25]. Conversely, Eagle et al. documented a sustained PAE in a mouse model in the apparent absence of any residual drug [13]. The reasons for these discrepancies are not clear. Eagle and Musselman showed that only actively growing organisms exhibited a PAE in vitro [12]. Pneumococci in CSF in vivo grow considerably more slowly than in broth, with generation times of 60 min and 20 min, respectively [29]. It is questionable, though, whether this difference in growth rate is the main reason for the absence of a drug-independent PAE in vivo since our in vitro studies showed no influence of the prolonged generation time in ex vivo CSF on the duration of the PAE.

Another interesting fact is that a PAE was observed in immunocompetent animals by Eagle et al. [13], whereas no drug-independent PAE could be documented in immunodeficient animals (neutropenic mice) [26] or in animals infected at a site of impaired host defenses (meningitis). It is conceivable that the occurrence of a PAE in vivo with *S pneumoniae* after exposure to penicillin is dependent on either the presence of residual amounts of drug or functioning host-defense mechanisms.

The relevance of a PAE to therapy is difficult to assess unequivocally. The pharmacologic parameter best suited to reflect the influence of the PAE is the interval between doses. However, when this interval is varied so that its influence on outcome

can be assessed, at least one other variable (the number of doses, the dose administered in a single injection, the total dose, or the duration of treatment) must also be changed. In our study we compared two different intervals (4 hr and 12 hr), varying the single and total doses of ampicillin while keeping the total duration of therapy constant (12 hr).

The most important factor determining efficacy of treatment in our experiments was the peak concentration of ampicillin in CSF. The outcome of treatment improved as drug concentrations in CSF increased, and peak concentrations that were >10 times the MBC consistently resulted in sterile CSF in >90% of animals. On the other hand, within the limits of the dosing schedules examined, the interval between doses and the duration of CSF ampicillin concentrations above the MIC did not appear to be of major importance with regard to outcome. If anything, prolonging of the interval between doses (with a constant amount of total drug) resulted in a favorable trend throughout all treatment groups; this effect was probably due to the resulting higher peak concentrations in CSF. Thus, even with small doses of drug, the benefit of the enhanced killing rate by the higher peak concentrations of ampicillin in CSF [30] seems to outweigh the hazard of eventual bacterial regrowth toward the end of the dosing interval. With large doses the total antibiotic effect observed in our preliminary experiments is apparently of sufficient duration to prevent bacterial regrowth during the 12-hr interval between doses.

Our findings are in contrast to those of Eagle et al. [13, 23, 24] and Gudmundsson et al. [26], which suggested that the duration of drug concentrations above the MIC was the most important determinant of outcome and that peak drug concentrations were of only minor import. It is possible that the unique characteristics of meningitis (i.e., bacterial growth conditions in CSF, pharmacokinetics in CSF, impaired host defenses in the subarachnoid space) are responsible for these discrepancies. Since the observed PAE in meningitis appears to be the result of a prolonged gamma phase of ampicillin elimination, direct extrapolation to infections at sites with different pharmacokinetics is not possible. Also, since the PAE may vary considerably from one antibiotic to another and from one organism to another [22], our results apply only to the drug and organism tested.

Our findings emphasize points of potential importance in the treatment of pneumococcal meningitis. If it is to effect maximal bacterial killing, the dose of drug administered must result in peak CSF concentrations far above the MBC for the pathogen. This notion has been confirmed in a recent study of the relation between the concentrations of several β-lactam antibiotics in CSF and the bactericidal effect of these drugs [30] and has also been supported by clinical experiences [5, 31, 32]. In addition, the drug-dependent PAE in CSF prevents immediate bacterial regrowth after drug concentrations drop below the MIC. The complexity of our model dictated the inclusion of only relatively small numbers of animals in each treatment group. The large inherent β-error that results from these small numbers precludes a definite statement about equality of the two intervals between doses. Nevertheless, our study demonstrates that the prolonged prevention of bacterial regrowth observed in vivo in CSF is associated with therapeutic efficacy. Furthermore, contrary to findings by Schmidt and Walley in experiments with a rat model of pneumococcal pneumonia [33], our studies failed to demonstrate a clear-cut advantage of long intervals between doses of drug. Thus, while our data suggest that an increase in these intervals can result in rates of cure similar to those obtained with sustained, high concentrations of drug in CSF as long as the peak concentration reaches a critical value (≥10 times the MBC), they do not support the concept that a prolonged therapeutic interval is superior.

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