CLINICAL MICROBIOLOGY REVIEWS, Apr. 1997, p. 345–357 Vol. 10, No. 2
0893-8512/97/$04.00+0

Management of Listeriosis

H. HOF,* T. NICHTERLEIN, AND M. KRETSCHMAR

Institute of Medical Microbiology and Hygiene, Faculty of Clinical Medicine Mannheim, University of Heidelberg, 68167 Mannheim, Germany

INTRODUCTION

Food-borne transmission of *Listeria monocytogenes* is the main route of acquisition of listeriosis (2). Hence, it is logical to augment efforts to prevent this disease by control of production and handling of food items. Furthermore, consumers should be continuously and broadly informed regarding the risks. There are several recommendations (2) that should be particularly stringently adhered to by susceptible persons, i.e., immunocompromised individuals and pregnant women. It should be kept in mind, however, that there are many other possible ways to acquire *Listeria monocytogenes.*

In spite of a potentially high risk of being exposed to these facultatively pathogenic bacteria in everyday situations, the rate of infection and the chance of developing overt disease is rather low (27). It is possible that most contacts will induce merely transient colonization in immunocompetent individuals or, at most, a mild disease that is generally so noncharacteristic that diagnostic tests are neglected. Consequently, such cases do not need specific antibiotic intervention. On the other hand, in patients with overt disease, the course of infection is often severe and even fatal. Overall, up to 30% of patients with manifest listeriosis die, clearly demonstrating that studies to further amend the therapy of listeriosis are an urgent priority.

* Corresponding author. Mailing address: Institute of Medical Microbiology and Hygiene, Klinikum Mannheim, Theodor-Kutzer-Ufer, 68167 Mannheim, Germany. Phone: 0621/383 2224. Fax: 0621/383 3816.

At first glance, the high lethality of listeriosis is difficult to understand, since virtually all strains of *L. monocytogenes* are susceptible to nearly all common antibiotics. Obviously, therapeutic success is impeded by the particular biological behavior of these bacteria, which are facultative intracellular bacteria (124). This means that in certain situations, the bacteria may be concealed from the extracellular environment that contains high concentrations of the antibacterial agents. Consequently, chemotherapy of listeriosis is complicated and requires extensive knowledge about the properties of the antibiotics as well as of the bacteria.

IN VITRO SUSCEPTIBILITY

Inhibition of Growth

Generally, isolates of *L. monocytogenes* are susceptible to a wide range of antibiotics (24, 50, 57, 109, 127) except for the newer cephalosporins and fosfomycin (Table 1). Other *Listeria* spp. behave similarly. It is noteworthy that most isolates from clinical as well as environmental sources are uniformly susceptible to these antibiotics; generally, a narrow range of MICs is found (Table 1). Thus, one can predict the susceptibility to antibiotics of a specific isolate with a high probability, which means that susceptibility testing is not mandatory in every case.

Resistance to certain antibiotics, such as streptomycin and kanamycin, which do not play a role in therapy, and erythromycin, gentamicin, trimethoprim, and rifampin, has been detected only in a few isolates from food (15, 25). The same is

TABLE 1. In vitro susceptibility of *L. monocytogenes* to various antibiotics[a]

Agent	MIC range (mg/liter)	Interpretation of inhibitory action			Interpretation of bactericidal action		
		Susceptible	Intermediate	Resistant	Yes	Tolerant or weak	No
Penicillin	0.06–2	•				•	
Ampicillin-amoxicillin	0.06–0.5	•				•	
Azlocillin	0.5–2	•				•	
Cephalothin	0.25–16		•				•
Cefotaxime	4–>128			•			•
Cefepime	4–>64			•			•
Imipenem	0.03–0.12	•				•	
Gentamicin	0.06–4	•			•		
Sisomicin	0.01–0.12	•			•		
Netilmicin	0.06–32	•			•		
Amikacin	<0.06–32	•			•		
Kanamycin	0.5–2	•			•		
Streptomycin	0.5–4	•			•		
Nalidixic acid	>128			•			•
D-Ofloxacin	>64			•			•
Levofloxacin	1–4		•				•
Ciprofloxacin	0.5–2		•				•
CI 934	0.25–1	•				•	
Bay Y 3118	0.06–0.25	•				•	
Vancomycin	0.12–0.5	•			•		
Teicoplanin	0.12–0.5	•			•		
Daptomycin	0.25–4		•				•
Co-trimoxazole	0.06–0.5	•			•		
Chloramphenicol	2–8		•				•
Rifampin	0.04–0.25	•					•
Fosfomycin	4–2,048			•			•
Clindamycin	0.25–4		•				•
Tetracycline	0.12–0.5	•					•

[a] Data from references 24, 42, 46, 50, 70, 86, 90, 109, and 119.

true for clinical isolates, in which resistance to streptomycin (25) or to tetracycline, chloramphenicol, and erythromycin (36, 103, 106) has been detected rarely. This antibiotic resistance was encoded by plasmids in most cases, although chromosomally encoded resistance to tetracycline has also been suggested (127). Since enterococcal plasmids encoding resistance determinants are easily transferred in vitro to *Listeria* spp., one could imagine that such a plasmid transfer might take place in nature much more often, for example, in the stools of humans and animals (17). It can be anticipated that this current favorable situation of uniform susceptibility to the antibiotics used in clinical practice might change under selective pressure during antibiotic treatment, particularly when drugs are used indiscriminately, as is often done in animal breeding.

It should be pointed out that resistance to penicillin derivatives has not yet been found under natural conditions (15, 57). Two reports on ampicillin-resistant cases are inconclusive, because they give no data on in vitro susceptibility testing. The conclusions are based only on clinical therapeutic failures (100, 108). Penicillin and ampicillin (75, 84, 138) are effective for the therapy of listeriosis. Amoxicillin has only a minor advantage over ampicillin (119). Acylureidopenicillins such as azlocillin, mezlocillin, and piperacillin are less active (24, 34, 75, 119), as are carbenicillin and ticarcillin (77, 119). The MIC of imipenem is even lower than that of ampicillin (22, 34, 61, 140),

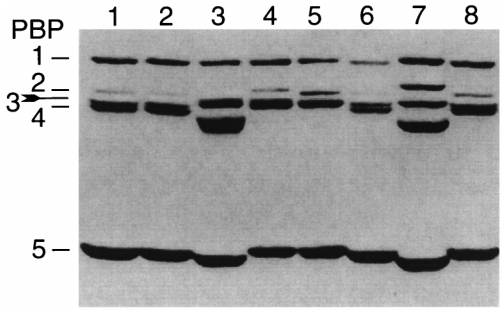

FIG. 1. PBP profiles of *Listeria* spp. Membranes of various *Listeria* spp. were incubated with radioactively labeled ampicillin and processed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis. Lanes: 1, *L. monocytogenes* Sv 1/2a SLCC 5779 (rough); 2, *L. monocytogenes* Sv 1/2a SLCC 2371 (NCTC 7973); 3, *L. welshimeri*; 4, *L. seeligeri*; 5, *L. ivanovii*; 6, *L. monocytogenes* Sv 4b SLCC 4013, 7, *L. innocua*; 8, *L. monocytogenes* SLCC 5779 (rough). Reprinted with permission from reference 37.

and resistance to this drug has been described only in genetically altered bacterial strains (99).

In contrast, most *Listeria* strains show a high natural resistance to cephalosporins, especially to those that are broad spectrum such as cefotaxime, and to monobactams. In this aspect, *Listeria* spp. resemble enterococci. The reason for this resistance is the lack of appropriate penicillin binding proteins (PBP) in their cytoplasmic membrane. Listeriae possess five PBPs that are characterized by different molecular masses (Fig. 1); the pattern of PBPs is fairly characteristic for a given *Listeria* sp. Only PBP3 appears to be identical in all *Listeria* spp. (37). The primary target for active β-lactams is apparently PBP3, since blockage of this enzyme, which is involved in the last stage of peptidoglycan synthesis, has lethal consequences for the bacterial cell (136). On the other hand, cefotaxime and aztreonam do not bind with high affinity to this essential PBP whereas penicillin and ampicillin bind with high affinity. The same holds true for PBP5 (99, 136). The affinity of the various β-lactams to the other PBPs is variable; the blocking of these enzymes is not immediately deleterious for the bacterial cell. The estimated copy number of the different PBPs per cell is low, i.e., PBP1, 120; PBP2, 80; PBP3, 145; PBP4, 130; and PBP5, 600 (136). At least a certain portion of these PBPs must be blocked before an antibacterial effect occurs.

In addition to the β-lactam antibiotics, the macrolide-lincosamide-streptogramin group of antibiotics has been of much interest, especially the macrolides, which are active with very few exceptions (103, 106) (Table 2). There is some confusion about the newer derivatives, because the MICs reported in the literature differ considerably (1, 8, 68). Possibly, clarithromycin is somewhat more active than erythromycin whereas roxithromycin is as active as erythromycin. Azithromycin and especially spiramycin are less active than erythromycin (1, 8). Among the

TABLE 2. In vitro activity of macrolides against *L. monocytogenes*

Agent	Activity obtained by:		
	Loza et al. (68)	Acar and Goldstein (1)	Bauer et al. (8)
Azithromycin		1–2	0.12
Clarithromycin	0.06–0.5	0.25–1	0.06
Erythromycin	0.1–1	0.25–1	0.25
Roxithromycin	0.2–1	0.50–1	0.12
Spiramycin		4	1

TABLE 3. Effect of a streptogramin (RP 59500) on *L. monocytogenes*

Strain	MIC (mg/liter)[b]	
	Erythromycin	RP 59500
EGD SLCC 5835	0.25	0.62
EGD pERL-3	>128	1.9
Courtieu[a]	>128	0.45

[a] Obtained from A. L. Courtieu (identical to the strain described in reference 103).
[b] Data from reference 91.

TABLE 4. MICs of sulfamethoxazole, trimethoprim, and co-trimoxazole for *L. monocytogenes*

Drug	MIC (mg/liter) for:		
	Strain SLCC 4013[a]	Undefined strain (142)[b]	Seven strains (13)[a]
Sulfamethoxazole	32	9.5	32–64
Trimethoprim	0.25	0.3	0.12–0.25
Co-trimoxazole	0.125	0.15	0.03–0.06

[a] Tested in Mueller-Hinton broth.
[b] Tested in tryptic soy broth.

lincosamide subgroup of agents, clindamycin is the most active drug against *Listeria* spp. (Table 1) (70). Cross-resistance exists between macrolides and clindamycin. The streptogramins (such as the pristinamycin derivative RP59500 [1, 91], which is a mixture of two similar derivatives, quinupristin and dalfopristin) are active in vitro, even against erythromycin-resistant strains (Table 3).

The aminoglycosides, particularly gentamicin, netilmicin, and amikacin, are equally effective (70) (Table 1). Chloramphenicol is only slightly active in vitro (Table 1) (70, 109).

From the beginning of the chemotherapy of listeriosis, the tetracyclines have been of particular value. Resistant strains which have acquired either plasmids or transposons (15, 104) that mediate the resistance have been isolated (15, 36, 103, 104, 106). Also, spontaneous development of tetracycline-resistant variants of *L. monocytogenes* has been reported (118). It is hoped that new derivatives such as glycylcyclines will be more active than tetracycline (130).

The quinolones do not yet play a key role in the treatment of listeriosis, since most of these agents have high activities against gram-negative bacteria but only moderate activities against gram-positive bacteria including *Listeria* spp. (52, 81, 109) and since highly resistant variants of *Listeria* have been selected during exposure to these drugs in vitro (13). Nalidixic acid, in particular, is completely inactive, and so it has been proposed as an additive to selective media for isolating *Listeria* spp. from polymicrobially contaminated sources (10). Ofloxacin is a mixture of two compounds, i.e., D-ofloxacin and levofloxacin. Whereas D-ofloxacin is completely inactive against *Listeria* spp., levofloxacin is moderately active (Table 1). Newer quinolone derivatives such as CI 934 (47) or Bay Y 3118 (90) exert high anti-listeria activities (Table 1). In addition, Bay Y 3118 has a pronounced postantibiotic effect (90). Like quinolones, coumermycin and novobiocin inhibit bacterial gyrase, although these agents belong to different chemical groups. Coumermycin is highly active against *Listeria* spp. (44).

A promising drug is rifampin, which is active in vitro (Table 1) (13, 24, 51, 81, 133). Resistant variants emerge under the influence of this drug, albeit at a low frequency of 10^{-7} (13).

The combination of trimethoprim with a sulfonamide such as sulfamethoxazole, as in co-trimoxazole, has been regarded as a drug of second choice for the treatment of listeriosis, because in vitro the MICs are rather low (Table 1) (13, 142). The major active agent seems to be trimethoprim, which is supported somewhat by sulfamethoxazole (Table 4). Resistance to trimethoprim has been reported only once (15).

The glycopeptides, such as vancomycin and teicoplanin or decaplanin (11, 42, 70, 86, 110, 134), as well as the lipopeptides, such as daptomycin (21, 46, 134), are active against virtually all isolates (Table 1). Tyrothricin, a cyclic peptide antibiotic consisting of gramicidin S and tyrocidin (123), displays considerable activity against listeriae. (The MIC determined by an agar dilution method on Mueller-Hinton agar was 1.25 to 2.5 mg/liter [93].) Fosfomycin is inactive against *L. monocytogenes* (24, 119) (Table 1).

Bactericidal Activities of Antibiotics

As noted above, most common antibiotics exert an inhibitory effect on *Listeria* spp. but only few are bactericidal. The β-lactam antibiotics, in particular, are only bacteriostatic for most isolates (24, 70, 113, 119, 133, 140), since there is a large gap between MICs and MBCs, at least when susceptibility tests are read at 24 h. Consequently, listeriae should be considered tolerant to all β-lactam agents, even though killing can be achieved after very long exposure (140). This is true for the majority (about 90%) of isolates, especially when bacteria are tested in the stationary phase. Furthermore, for most strains, the so-called Eagle effect can be observed, meaning that with increased β-lactam concentrations, the MBCs increase also (120, 133). The reason for this particular behavior of listeriae remains obscure. To explain this phenomenon, investigations of peptidoglycan synthesis and regulation of the autolytic system are still needed. The importance of the structure of the cell wall of listeriae (26) is not yet known. Optimal killing by β-lactams is achieved only when several of the different PBPs are blocked and when a large proportion of each PBP is saturated (112).

Bactericidal activities have been seen with aminoglycosides (129, 133), teicoplanin (11), vancomycin (13, 42, 70), and co-trimoxazole, in which trimethoprim plays the prominent role (133, 142). These agents differ, however, in the speed of killing: aminoglycosides have accomplished this task within 1 to 2 h whereas vancomycin and co-trimoxazole need 6 to 24 h, respectively. Although these findings have been confirmed many times, individual isolates may be more resistant than most to the bactericidal action of a certain drug, for example to the aminoglycosides (133).

Tyrothricin is rapidly bactericidal for listeriae, killing 99.9% of the inoculum in 1 min (Fig. 2A). These drugs are bactericidal because they produce channels in the membrane, which rapidly results in the death of target cells (123). Rifampin is not bactericidal for listeriae (113, 133, 140). The killing capacity of quinolones in particular for *Listeria* spp. is only weakly expressed (47, 52, 90) (Fig. 2B). A reduction of bacterial counts is achieved only at concentrations far beyond the MICs. Killing occurs rather late, i.e., several hours after exposure to the antimicrobial agent, whereas with gram-negative bacteria, killing occurs rapidly, often within an hour. Coumermycin, another gyrase inhibitor, also is not bactericidal for listeriae (44).

The agents that are considered bacteriostatic in general, such as macrolides, tetracyclines, and chloramphenicol, are bacteriostatic for listeriae as well.

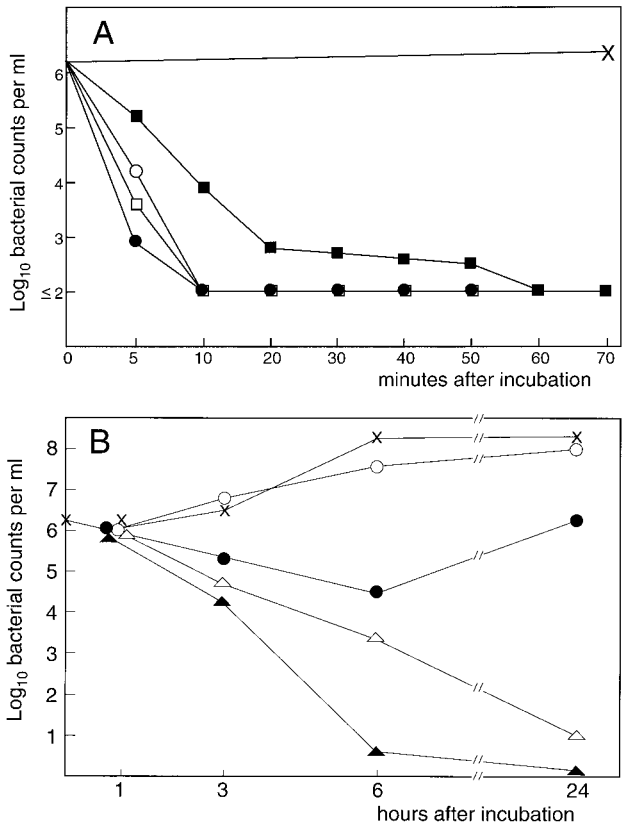

FIG. 2. Bactericidal activities of antibiotics against *L. monocytogenes* incubated in vitro in Mueller-Hinton broth. (A) Tyrothricin. Symbols: X, untreated control; ■, MIC = 2.5 mg/liter; ○, MIC = 5 mg/liter; □, MIC = 10 mg/liter; ●, MIC = 20 mg/liter. (B) CI 934, a quinolone. Symbols: X, untreated control; ○, MIC = 0.125 mg/liter; ●, MIC = 0.5 mg/liter; △, MIC = 2.0 mg/liter; ▲, MIC = 8 mg/liter. Panel A reprinted with permission from reference 63; panel B reprinted with permission from reference 47.

Effects of Antibiotic Concentrations below the MIC

In vivo, at remote sites of infection, such as the intracellular environment where listeriae may reside, the concentration of active antibiotic is presumably not consistently above the MIC during the entire therapy. Therefore, the effect of subinhibitory concentrations of antibiotics on listeriae is of interest.

Ciprofloxacin at concentrations below the MIC induces a marked morphological change in *Listeria* spp. within 3 h (52). Long filaments are produced, and these bacteria resemble *Listeria* cells lacking p60, the product of the *iap* gene, which is an essential virulence factor (102, 124). This appearance might indicate reduced production of this cell wall protein under antibiotic treatment.

Another target of antibiotic concentrations below the MIC is the hemolysin (listeriolysin), a major virulence factor of *L. monocytogenes*. Modulation of listeriolysin production by antibiotics was measured by a hemolysin assay with human erythrocytes and measurement of β-galactosidase expressed from a *lacZ* fusion of the listeriolysin promoter (93). Ampicillin reduced the production of listeriolysin and β-galactosidase when tested at subinhibitory concentrations that did not affect the growth of the listeriae. Broad-spectrum cephalosporins, although devoid of inhibitory activity against listeriae, are also able to reduce the expression of this essential virulence factor (93), as are vancomycin, DL-cycloserine, fosfomycin, tyrothricin, gentamicin, and doxycycline but not rifampin, fusidic acid, or gyrase inhibitors (93). Notably, this reduction is not accompanied by a general metabolic inhibition, since expression of the β-galactosidase gene under the regulation of a streptococcal promoter is unaltered.

Synergism or Antagonism of Antibiotic Combinations

Since the β-lactams are bacteriostatic for listeriae only in vitro, additional antibiotics are needed for a bactericidal effect. For this purpose, their use in combination with aminoglycosides is usually recommended. True synergism has been found with penicillin plus gentamicin (83) as well as ampicillin or amoxicillin plus gentamicin (4, 13, 31, 82, 119, 129, 139). The combination of imipenem with gentamicin is equally synergistic (22, 61). The consequence of the simultaneous action of both groups of antibiotics is a better and more rapid killing effect; bacteria begin to die after 30 min (129), even at concentrations of each agent that are only bacteriostatic when tested alone (83).

On the other hand, the combination of ampicillin with rifampin is only additive (133) or even antagonistic (13, 113, 140). The combination of penicillin or ampicillin with erythromycin is antagonistic (98), and chloramphenicol interferes with the antibacterial effect of ampicillin (140). Other drug combinations do not show any beneficial effect (69).

An observation that killing of listeriae by ampicillin might be enhanced by lysozyme (3) may be closer to the in vivo situation.

Susceptibility to Bacteriocins and Lantibiotics

In their natural habitat, for example in foods and feces of animals and humans, listeriae may be regularly exposed to antibacterial agents produced by other bacterial members of the microflora. Bacteriocins are antibiotic peptides with a rather narrow spectrum of activity. In general, they are produced by bacterial species that are closely related to the target species. The producer strain, however, has developed a protective mechanism against its particular agent. The bacteriocins that are active against gram-positive bacteria generally have a low molecular mass, i.e., not more than 10,000 Da, and have a broader spectrum of activity than do bacteriocins that are active against gram-negative bacteria. Among the bacteriocins of gram-positive bacteria, a particular group can be separated, the lantibiotics (pep5, subtilin, epidermin, gallidermin, nisin, and mersacidin), which are characterized by the presence of an intramolecular ring moiety that contains lanthionine, a peculiar amino acid. These peptides diffuse across the wall of gram-positive bacteria and depolarize the cell membrane of the target organism, for example by producing channels and thus causing efflux of ions and small molecules (55, 56).

Various bacterial species of the microbial flora of foods and feces may be able to synthesize bacteriocins. The *Clostridium-Lactobacillus-Bacillus* branch of gram-positive bacteria is closely related to *Listeria* spp. and may play a role in the regulation of growth of *Listeria* spp. (85). Meat contaminated with *L. monocytogenes* can be effectively cured by addition of a *Lactobacillus* sp. producing a bacteriocin (116).

Brochocin-C, a bacteriocin produced by *Brochotrix campestris*, a species closely related to *Listeria* spp., is highly active against the latter. Smooth and rough strains of *L. monocytogenes* are both susceptible (125).

Leuconostoc carnosum (115, 135) and *Leuconostoc mesenteroides* (38) also produce bacteriocins that act against *Listeria* spp. Among 20 strains tested, 3 had activity against *Listeria* spp. and other gram-positive bacteria, including *Lactobacillus* and *Enterococcus* spp. (135). Killing of 99.9% of the inoculum was

achieved within 1 min. All *Listeria* strains tested so far are susceptible to this bacteriocin to various degrees. This agent is stable over a wide range of pH but labile to the action of proteases (135). The bacteriocin PA-1 from *Pediococcus acidilactici* is able to reduce the listerial load of cheese at various temperatures, even in the refrigerator (105).

Interestingly, other bacterial species, such as enterococci, may also produce bacteriocins with antilisterial activity (96). Thus, it may be speculated that such a mechanism could influence the colonization of the intestine with *L. monocytogenes* and that bacteriocins may help to prevent infection after consumption of contaminated food. The lantibiotic mersacidin, produced by *Bacillus* spp., which is active against a wide range of gram-positive bacteria, has only moderate activity against *L. monocytogenes* in vitro (MIC, 4 to 16 mg/liter) (63). Whereas this agent has been shown to be bactericidal in vivo against other gram-positive bacteria and to be even more effective than vancomycin, its activity against *L. monocytogenes* is only bacteriostatic. In cell cultures and in mice, this agent is inactive against intracellular *L. monocytogenes* (63). Gallidermin, another lantibiotic, also has good activity in vitro against *L. monocytogenes* and other *Listeria* spp. (29).

The concept of using bacteriophages as antimicrobial agents has generally been unsuccessful in practice. An interesting modern approach is to use specific gene products of bacteriophages, such as lysin, an enzyme produced by lytic bacteriophages, to rupture the cell wall of a host bacterium. This product is able to destroy *L. monocytogenes* and other *Listeria* spp. (97).

Susceptibility to "Endogenous Antibiotics"

Mammals, insects, and plants produce a series of antimicrobial compounds that regulate the colonization, entry, and replication of bacteria within the host. Such "endogenous antibiotics," such as magainins, cryptdins, defensins, and cationic proteins, are responsible for a very potent, nonspecific humoral defense against a variety of microorganisms, including listeriae (23, 33, 41, 65). Some of these products are stored within host cells, for example in the granules of polymorphonuclear leukocytes and macrophages, whereas others are secreted into internal or external fluids. In principle, their activities might be of biological relevance for *Listeria* infection, by counteracting the colonization of mucosal surfaces with pathogenic listeriae and inhibiting their intracellular multiplication. *L. monocytogenes* is likely to be inhibited by some of these compounds (122).

ACTIVITY OF ANTIBIOTICS ON *L. MONOCYTOGENES* RESIDING WITHIN HOST CELLS

Specific Intracellular Site of Replication

The virulence of *L. monocytogenes* strains is specifically characterized by their property of penetrating actively into a wide range of host cells; not only professional phagocytes but also epithelial and parenchymal cells may be invaded by either phagocytosis or induced phagocytosis (124). A cluster of virulence genes enables pathogenic *Listeria* strains to evade the phagocytic vacuole and to reside and multiply within the cytoplasm. Shortly after reaching the cytoplasm, the bacteria are wrapped by a sheet of host cell-derived actin filaments, which form a comet-like structure at the back pole of the bacterium. The bacterium is seen to move across the cytoplasm at a rather high speed, i.e., 1 μm/s. Occasionally, the bacterium gets beyond the membrane of the host cells, which are triggered to produce long protrusions enclosing the *L. monocytogenes* cells. These cell protrusions are then internalized by an adjacent host cell (131). This phenomenon of direct cell-to-cell spread means that the bacteria are hidden within host cells during the course of infection and therefore are protected from the humoral defense mechanisms of the host and from antibiotics in the extracellular fluids. Listeriae have the potential to invade and replicate in many different cells of mesenchymal or epithelial origin. However, quantitative differences in the rates of multiplication may influence the outcome of therapy, such as the effectiveness of treatment with β-lactam antibiotics.

TABLE 5. Antibiotic uptake by eukaryotic host cells and intracellular distribution of antibiotics.

Drug(s)[a]	Intracellular distribution in[b]:	
	Cytoplasm	Lysosomes
Group I (ratio, >10)		
Macrolides (erythromycin, roxithromycin, azithromycin, spiramycin)	+	+
Clindamycin	+	+
Rifapentine	+	−
Coumermycin	NA	NA
Streptogramin (RP 59500)	NA	NA
Group II (ratio, 1–10)		
Quinolones (ofloxacin, ciprofloxacin, CI 934)	+	(+)
Rifampin	+	+
Tetracycline	NA	NA
Chloramphenicol	NA	NA
Vancomycin	NA	NA
Teicoplanin	NA	NA
Fosfomycin	NA	NA
Group III (ratio, <1)		
Penicillin	+	−
Ampicillin	+	−
Cephalosporins	+	−
Imipenem	+	−
Aminoglycosides (gentamicin, netilmicin, amikacin)	−	+

[a] Intracellular/extracellular ratio.
[b] +, strong accumulation; (+), minor accumulation; −, no accumulation; NA, no data available.

Uptake and Intracellular Distribution of Antibiotics

During listeriosis, a therapeutic effect of an antibiotic treatment is not dependent only on the direct inhibitory or bactericidal activity of an agent. Because of the intracellular habitat of *L. monocytogenes* several other prerequisites must be fulfilled.

(i) A drug must be taken up into the host cell either by passive diffusion or, preferably, by active transport. In this regard, there are large differences among the common antibiotics active against listeriae (Table 5) (49). It is worth noting, however, that the results obtained with one cell type cannot be extrapolated directly to other cell types. Certain host cells may be substantially devoid of antibiotics if they have special transport systems in their cytoplasmic membranes, which rapidly export certain infiltrated antibiotics (92). Furthermore, the amount of internalized antibiotic may depend on the physiologic state of a host cell; for example, a cell that is already burdened with intracellular bacteria may have impaired antibiotic uptake (49).

(ii) Within a host cell, certain drugs, such as azithromycin, are tightly bound to intracellular targets and thus create depots whereas others, such as quinolones, easily diffuse out of the host cell when the extracellular concentrations decrease (49).

(iii) The mere presence of an antibiotic within a host cell does not guarantee therapeutic success. Some antimicrobial agents gain access to an improper compartment in the host cell and thus cannot interact with the bacterial invader. For example, teicoplanin remains adherent to the membrane of the host cell after penetration; gentamicin is exclusively transported into the lysosomes and has no contact with intracellular listeriae there. The quinolones and the macrolides are accumulated within the lysosomes and the cytosol, whereas the β-lactams generally cross the cell membrane in small amounts only; thereafter, they are located mainly in the cytosol (Table 5).

(iv) The environmental conditions within an intracellular compartment, such as protein binding, ionic strength, and pH value, do not allow an optimal antimicrobial effect for some drugs. For example, at the low pH of the lysosomes, gentamicin is present only in its protonic isoform, which is completely inactive (62).

(v) Notably, unrelated drugs that do not possess antimicrobial activity but are used concomitantly with antibiotics can influence their intracellular accumulation, distribution, and activity. For example, probenecid enhances the accumulation of quinolones within eukaryotic cells (14). Chloroquine and amantadine are transported to the lysosomes of a host cell, where they elevate the pH of these intracellular compartments and hence interfere with the activity of intracellular antimicrobial agents such as tetracycline (107).

Thus, although listeriae are susceptible in vitro to most of the common antibiotics, many possibilities for therapeutic failures exist. In addition, it is probable that determination of MICs and MBCs in vitro on artificial media is not sufficient to predict the therapeutic value of a drug in the treatment of listeriosis.

Intracellular Activity of Antibiotics on *L. monocytogenes*

The above theoretical reflections lend support to doubt whether conventional susceptibility testing is sufficient for ranking drugs used for therapy of listeriosis. Thus, for laboratory evaluation of the drugs, cell cultures have been infected with virulent *L. monocytogenes* and treated with antibiotics. There are, however, considerable methodological and interpretive difficulties with these tests.

Elimination of extracellular bacteria. A technical problem impedes the correct interpretation of results obtained with cell cultures. Only a small portion of added listeriae is taken up avidly by eukaryotic cells; the remaining extracellular bacteria multiply in the cell culture medium and could obscure the effect of an antibiotic on intracellular listeriae. Thus, in most such experiments, a rapidly bactericidal agent is incorporated at relatively high concentration into the extracellular medium to eliminate virtually all noninternalized bacteria. In most instances, gentamicin is used for this purpose, assuming that intracellular bacteria, as is the case with *Salmonella typhimurium*, for example (60), are not affected by this drug.

This assumption, however, has been challenged recently (18). The authors claim that at least in professional phagocytes, extracellular fluid containing gentamicin may gain access to intraphagosomal listeriae and kill the internalized bacteria before they can enter the cytoplasmic space. In most experiments with nonprofessional phagocytes, such as L929 fibroblasts or HeLa cells, this mechanism is unlikely to influence the results, because the listeriae grow readily in the treated cells. Furthermore, gentamicin is not held to be inactive against intracellular bacteria because of failure of penetration. Indeed, gentamicin is taken up to a certain extent depending on the time of exposure to the drug but is stored primarily within lysosomes (62). After fusion of lysosomes with a phagocytic vacuole, this agent may have contact with internalized bacteria, but in normal, acidified vacuoles, the pH is highly detrimental for gentamicin, which is transformed into its antimicrobially inactive protonic form (62). Thus, one can conclude that gentamicin is well suited for the rapid elimination of extracellular listeriae without influencing either intravacuolar or intracytoplasmic listeriae.

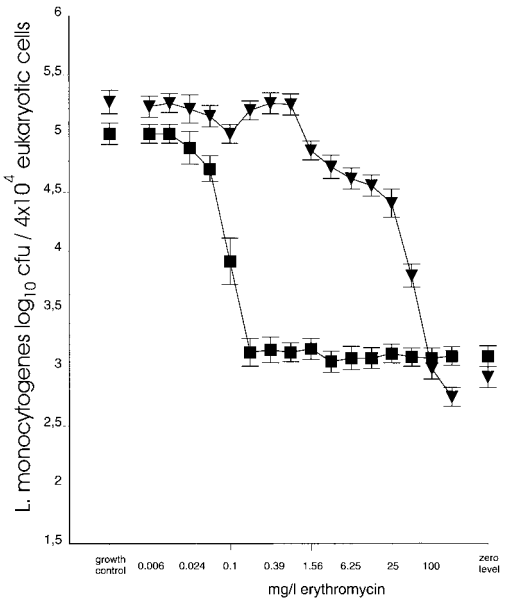

FIG. 3. Activity of erythromycin on intracellular *L. monocytogenes* (MIC = 0.25 mg/liter) in normal (KB3-1) (■) and MDR$^+$ (KBV-1) (▼) cells, respectively. Reprinted with permission from reference 92.

In future experiments, the elimination of extracellular listeriae may be attempted by treatment with bacteriophages, bacteriocins, or lantibiotics.

Proper host cell type. In complex situations such as those existing in an animal or human body, some cells may differ from other cells by their ability to take up and then dispose of internalized antibiotics by active export. Several such membrane transport systems, like multidrug resistance (MDR) (32) or multidrug resistance-associated protein (76), export antibiotics. There is evidence from cell culture experiments that *L. monocytogenes* organisms multiplying in cells with overexpression of MDR are protected against the action of erythromycin (Fig. 3). Whereas in normal KB3-1 cells, originally derived from HeLa cells, a concentration of 0.1 mg of erythromycin/liter or more in the supernatant fluid reduces the intracellular bacterial counts, in KBV-1 cells that overexpress MDR, a 1,000-fold-increased amount of drug is required for the same effect (92).

Therapeutic consequences. Penicillin, ampicillin, and amoxicillin can reduce the numbers of intracellular listeriae, indicating a bactericidal effect (81, 89). This effect is achieved at concentrations that may be obtained in the body during treatment with conventional amounts of drug. At first glance, this observation is astonishing because β-lactam antibiotics do not belong to a group of intracellularly accumulated antibiotics (49); the intracellular concentrations are comparatively lower

than the extracellular concentrations. Furthermore, it must be kept in mind that listeriae are tolerant to β-lactams in vitro, which means that they are killed only by high concentrations of the drugs. It might be speculated that the unexpected intracellular killing by β-lactams is due to a reduced expression of essential virulence factors by the pathogen even at the low concentrations of β-lactams, possibly even below the MIC, that are achieved in the cytosol of host cells. Thus, the killing of intracellular listeriae might result from host cell mechanisms rather than from antibiotics.

Ampicillin incorporated into liposomes is more effective against *L. monocytogenes* in mouse peritoneal macrophages than is ampicillin alone. The liposomes enhance antibiotic uptake (6). However, higher drug concentrations may not be achieved in other relevant host cells, such as hepatocytes, which also may be infected by *L. monocytogenes*. Apart from these β-lactams, co-trimoxazole also has a bactericidal effect on listeriae within host cells (81). Rifampin, erythromycin, doxycycline, and coumermycin inhibit multiplication rather than kill listeriae (81, 89).

Ciprofloxacin and chloramphenicol are not inhibitory at all. The newer quinolones sparfloxacin (81) and Bay Y 3118 (90) are more bactericidal than older formulations, which means that at concentrations about the MICs, a reduction of intracellular counts is seen.

The newer macrolides such as clarithromycin are superior to erythromycin in that they inhibit the growth of listeriae at lower concentrations, but they are not bactericidal for intracellular listeriae (88). As discussed above, the distribution and activity of intracellular antibiotics may be markedly influenced by environmental conditions or by drugs with no inherent antibacterial activity, such as chloroquine or amantadine. These agents are lysosomotropic and alkalinize lysosomal locations, which may result in a change in the activity of certain antibiotics (107). As a consequence, experimental results cannot always be extrapolated to the situation in the whole animal. Some cells that intrinsically express export pumps such as MDR (32) may not accumulate macrolides intracellularly. As MDR is expressed differently in various tissues (132), either intrinsically or induced by environmental conditions, one might speculate that the export of antibiotics by some infected cells could contribute to treatment failure and persistence of listeriae. MDR often is overexpressed in tumor cells (30), so that these host cells might provide a safe haven for *L. monocytogenes.*

THERAPEUTIC ACTIVITY OF ANTIBIOTICS IN ANIMAL MODELS

Systemic Infection of the Spleen and Liver in Mice—Intracellular Multiplication

A rough estimation of antibiotic activity can be obtained by determining the survival rates or survival times after inoculation of animals with high, lethal doses of *L. monocytogenes*. Subcutaneous or intraperitoneal injection of an appropriate (nonlethal) number of virulent listeriae into mice leads to a systemic infection which affects mainly the organs of the mononuclear phagocyte system, such as the spleen and liver, but does not induce meningitis or encephalitis. A well-defined infection curve can be constructed by counting the bacterial numbers in these organs several days after infection (50, 71). This technique allows precise and quantitative follow-up of bacterial multiplication. The effect of antibiotics given either orally or parenterally can thus be sensitively monitored. A general problem with the use of small animals is that the pharmacologic properties of antibiotics may differ substantially from those in humans and the dose and intervals of administration, which are often arbitrarily chosen in such animal models, hardly apply to human therapy.

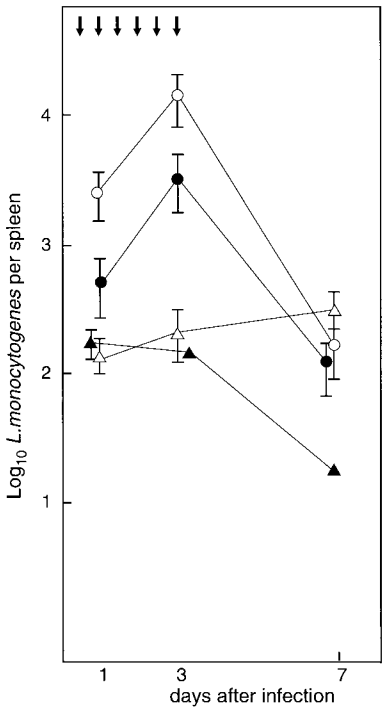

FIG. 4. Therapeutic activity of CI 934, a new quinolone, on murine listeriosis. Symbols: ○, untreated control mice; ●, 1 mg twice a day given orally; △, 1 mg twice a day given intraperitoneally; ▲, 1 mg twice a day given subcutaneously. Arrows indicate the times when antibiotics were administered. Reprinted with permission from reference 47.

A real advantage of this *Listeria* model is that therapy can be started several hours after infection and continued for several days. If therapy were begun immediately after injection of listeriae, as is done in most other animal models for evaluation of antibiotics, most listeriae would still be extracellular and easily attacked by the antibiotic. Several hours after infection, a typical inflammatory response characterized by a serous exudate, followed by formation of granulocytic abscesses and even later by organized granulomas, develops (40). These histologic alterations clearly influence the therapeutic outcome, as proven in a historic paper by Porter and Hale (101). They reported on the therapeutic effect of sulfonamides on *Listeria* infection of mice. Survival was enhanced when treatment was started early, i.e., 3 h after inoculation with listeriae. Since sulfonamides attack intracellular bacteria poorly, it could be assumed that these antimicrobial agents act on extracellular listeriae to reduce the challenge dose of infective organisms to a sublethal level. It is generally accepted that in later stages of infection, listeriae reside and multiply predominantly within macrophages and even within parenchymal cells of animals. Initiation of therapy 6 h after infection clearly allows the therapeutic effect of an antibiotic to concentrate on an established infectious process. In principle, three different aspects of therapy can be described.

Curing. Only a few agents, such as rifampin, coumermycin, the newer quinolones, CI 934 (Fig. 4) and Bay Y 3118 (47, 54, 90), and the newer macrolides, clarithromycin, azithromycin, and roxithromycin (8), are able to cure infection rapidly. Am-

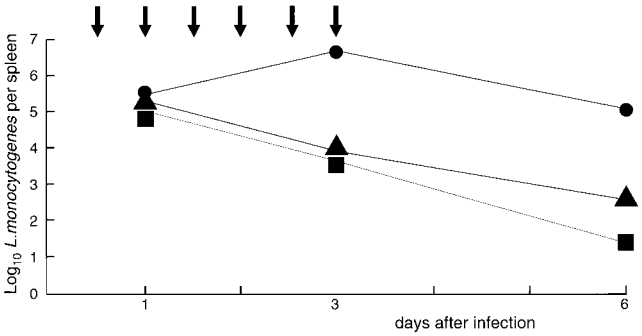

FIG. 5. Therapeutic activity of ciprofloxacin in comparison to ampicillin on murine listeriosis (90). Symbols: ●, untreated controls; ▲, 2 mg of ciprofloxacin twice a day given intraperitoneally; ■, 2 mg of ampicillin twice a day given intraperitoneally. Arrows indicate the times when antibiotics were administered.

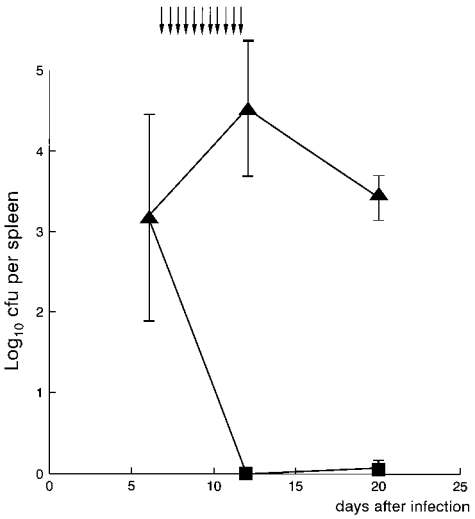

FIG. 6. Effect of 2 mg of Bay Y 3118, given twice a day, on listeriosis in immunocompromised, nude mice (■) (90). Untreated control mice (▲) could not eliminate the bacteria. Arrows indicate the times when antibiotics were administered.

picillin achieves this task rather late, only after a few days, and elimination is complete only when the immune response occurs and acts in conjunction with the antibiotic (Fig. 5) (54, 121). An improved response is seen after incorporation of ampicillin into liposomes (5), which are engulfed primarily by Kupffer cells of the liver. Protracted liberation of the drug, which protects hepatocytes, may be the reason for this better therapeutic effect, rather than simply an increased amount of drug in the Kupffer cells.

In the mouse model, the addition of gentamicin to ampicillin does not improve the therapeutic result (53), probably because the intracellular listeriae are not reached by gentamicin. Indeed, the bulk of listeriae reside within liver and spleen cells. On the other hand, the survival of mice infected with a high dose of *L. monocytogenes*, which leads to death within 2 days, was somewhat increased when penicillin was combined with gentamicin (20). This could be explained by the fact that in this model the excessive numbers of bacteria induce necrosis (40). In areas where the normal architecture of tissues is destroyed, most bacteria will lie extracellularly and hence are rendered accessible to gentamicin according to the in vitro situation allowing synergism with penicillin.

Inhibition. Another group of agents including imipenem, azlocillin, tetracyclines, erythromycin, spiramycin, co-trimoxazole, ciprofloxacin (Fig. 5) (8, 54), vancomycin, teicoplanin (11), and RP 59500 (91) inhibits the multiplication and dissemination of listeriae but does not reduce bacterial counts until the body's own defense mechanisms finally stop the infection.

Failure. A third group of antibiotics such as mezlocillin and cephalosporins (54) or mersacidin (63) has no effect on the course of infection, although mezlocillin and mersacidin are active in vitro.

Acute Meningitis in Rabbits—Extracellular Multiplication

In the rabbit model, acute meningitis is induced by intracisternal inoculation of a high dose ($>10^7$ cells) of virulent listeriae. A purulent infection develops rapidly, within 15 h. During this short-lived process, treatment can be given only over a very short period. In untreated animals, the course of infection is rapidly fatal (95, 113, 114). It may be assumed that in this situation, when viable bacteria in the cerebrospinal fluid (CSF) appear to be resistant to phagocytosis by polymorphonuclear leukocytes (111), the bulk of listeriae may lie extracellularly, as has been pointed out by Winslow et al. (140).

In this model, penicillin is highly beneficial and ampicillin (114) and mezlocillin (95) are even more so, whereas rifampin is less active, showing that the ability of a drug such as rifampin to accumulate within host cells does not afford an advantage over extracellular agents; there is no synergism of rifampin with penicillin (113). This may be analogous to the in vitro situation in which no synergism occurs. Another reason may be the poor penetration of rifampicin into the CSF (113).

In contrast, rifampin alone (51) or in combination with ampicillin (137) is highly active in the murine model of systemic infection in which intracellular multiplication of *L. monocytogenes* is predominant. Equally, in the murine model, the combination of ampicillin and gentamicin is indifferent (53), whereas in the meningitis model with largely extracellular bacteria this combination is more active than either drug alone (114), reflecting the in vitro situation (4, 13, 31, 82, 119, 129, 139).

Effect in Immunocompromised Mice

A quite different condition is created in a model of infection of congenitally athymic, nude mice. In these mice, a chronic course of infection is induced by virulent strains of *L. monocytogenes*. Consequently, treatment can be started several days after infection, when histological alterations have already developed (40, 50). In addition, this model approaches more closely the clinical situation in humans, because these animals are immunocompromised by the lack of T lymphocytes and are analogous to patients with leukemia or AIDS. In this situation, the role of an antibiotic is much more decisive, since the cooperation of the body's own defense mechanisms is lacking.

In immunocompromised nude mice that develop a chronic infection, the first dose of antibiotic can be given after infection is established and when inflammatory reactions in the infected tissues are present. Under these circumstances, the therapeutic effect of only very few agents remains satisfactory. Coumermycin (44, 54) and the new quinolone Bay Y 3118 (90) are still curative (Fig. 6). Rifampin and ampicillin (Fig. 7) are able to reduce the bacterial counts (51, 54) but not to cure the infection; thus, an exacerbation after cessation of antibiotic therapy occurs, because the few remaining bacteria cannot be elimi-

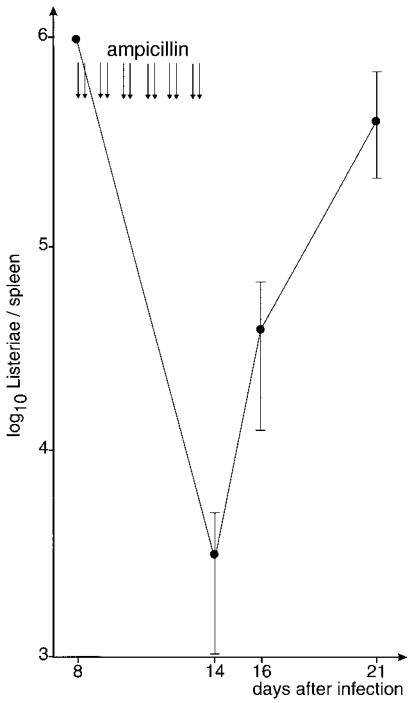

FIG. 7. Effect of 2 mg of ampicillin on listeriosis in immunocompromised, nude mice. Arrows indicate times of antibiotic administration. Reprinted with permission from reference 50.

nated by the host's defective defense system. Even liposome-entrapped ampicillin is not protective (5). Bacteriostatic agents, such as erythromycin or tetracycline, do not show any therapeutic effect in these compromised animals (54).

IMMUNIZATION AND IMMUNOMODULATION

Since resistance to infection with *L. monocytogenes* is cell mediated (71), killed vaccines are not protective. The best protection can be achieved by infection with a virulent strain that multiplies in the host, but even this effect fades away within several months (43). The use of attenuated vaccine strains is hampered by the fact that to achieve protective immunity, an opportunity to multiply within host cells must remain. For immunocompromised hosts, there is the risk that this may lead to disease. One strategy is to immunize against certain virulence factors of *L. monocytogenes*, for example p60, a major protein of the bacterial surface. This listerial antigen, when expressed by other vectors, for example by avirulent strains of *Salmonella typhimurium*, is able to induce a certain degree of protection (28). However, this approach is far from being applicable for routine vaccination of animals or humans.

Another idea is to nonspecifically strengthen the body's own defense system. Training of the mononuclear phagocyte system by previous confrontation with nonspecific stimulants such as lipopolysaccharide, *Mycobacterium bovis* BCG, or killed *Bordetella pertussis* cells (143) helps to overcome infection in experimental animals but is not of practical use because of side effects. In principle, all such agents must be given at a definite interval before the experimental infection to influence the course of infection, because there is a lag phase between the stimulus and the reaction of the defense system. Such a protocol is unsuitable for practical purposes, because one cannot predict the exact time point of infection and the beneficial effects are only short-lived. Activation of the T-lymphocyte system through nonspecific modulators, such as cytokines, has been tested in animal experiments. For example, interferon confers some degree of resistance against infection with *L. monocytogenes* in normal mice (59) but not in nude mice (45). Obviously, one instrument alone is not able to replace a concert of diverse agents and factors.

Immunmodulation is not yet a real practice in medicine, although sometimes it may be an undesired side effect of antibiotics themselves, i.e., by direct alteration of granulocyte or lymphocyte function (80).

CLINICAL EXPERIENCE

Since listeriosis is a rather rare human disease which occurs sporadically or in small epidemics in most instances, prospective clinical studies on the best antibiotic regimen are not available. The present view of the optimal therapy is founded on occasional observations and impressions. Thus, in the literature one finds reports on limited numbers of patients treated with a certain regimen but no direct comparison of two different choices. Because the result of chemotherapy may also depend on the activity of the body's own defense system, which may differ in neonates, adults, and cancer patients, the contribution of antibiotics to a cure may be difficult to evaluate in each case. In addition, the clinical manifestations of disease, for example meningitis or encephalitis, may influence the outcome of the infection and the susceptibility to therapy.

Penicillin derivatives play a pivotal role in the therapy of all forms of listeriosis. Penicillin G given intravenously in high doses has been used with success for meningitis as well as other manifestations (79). With ampicillin, too, good results have been obtained in adults (138) and in neonates (72, 84). Therapeutic failures may not be due to the resistance of *L. monocytogenes* to ampicillin as stated previously (100, 108). In a direct comparison, the efficacy of ampicillin was superior to that of penicillin (64); however, this could not be confirmed in a larger study (94). The therapeutic importance of the acylureidopenicillins (74) or imipenem has not yet been clarified. In contrast, cephalosporins were completely ineffective (94). Listeriosis developed even in patients undergoing therapy with cephalothin (73) and cefazolin (66). Other cephalosporins, such as cefotaxime, ceftriaxone, and ceftazidime, are all clinically ineffective. However, it seems important to note that cephalosporins do not interfere with the action of the penicillins when added to the treatment regimen (16).

The effectiveness of chemotherapy depends largely on the dose administered. High doses of ampicillin are needed, especially when infection of the brain occurs, since ampicillin crosses the blood-brain barrier only poorly; not less than 6 g/day must be given to an adult (74), but much larger amounts have to be used in certain cases; for example, 18 g/day has been used in a pregnant woman antepartum (58).

Apart from the dosage, the duration of treatment seems to be crucial. In general, a period of at least 2 to 3 weeks has been advised to avoid relapse (74, 126). This holds true particularly for immunocompromised patients, who are often found among persons with listeriosis (67, 94) and in whom even a few remaining bacteria are not killed by the body's own defenses. Furthermore, it should be remembered that *L. monocytogenes* is tolerant to the action of ampicillin, so that the bactericidal activity of this antibiotic is not reliable (see above). Indeed, recurrent listeriosis with the same strain has been observed in some patients (78, 126).

In spite of rational antibiotic therapy with ampicillin, about one-third of patients with listeriosis die; therapeutic failure is

probably due not to the antimicrobial resistance of the pathogen but, rather, to the stage of the infectious disease or to the critical situation of the patient as a result of the underlying disease. Whereas previously healthy patients have a relatively good chance of surviving infection, those with underlying disease are at especial risk of succumbing to listeriosis (94, 126, 144).

From in vitro data (see above) and animal experiments (114), it has been determined that the addition of gentamicin to ampicillin could further enhance the therapeutic effect. Although there are no comprehensive clinical studies, substantial evidence has been derived from observations of many favorable outcomes when this regimen is used. Today, there is ample agreement that this combination is the therapy of primary choice for listeriosis (74). For patients allergic to β-lactams, co-trimoxazole has been recommended as the therapy of second choice (16, 128, 141) for intracranial as well as extracranial manifestations. Another favored aspect of co-trimoxazole is that after an initial parenteral application, therapy can be continued orally, for example with trimethoprim alone, to prevent relapse (35). Use of the combination of ampicillin and co-trimoxazole has also been recommended (9), although experience with this combination is rather limited. All other drugs, like chloramphenicol, tetracyclines, and macrolides, are much less potent (16, 64).

Contradictory results have been obtained with vancomycin. Although this drug has been recommended as an alternative to ampicillin or co-trimoxazole (12), treatment failures have been reported (19) and the development of *Listeria* meningitis during vancomycin therapy, possibly due to poor penetration of this drug into the CSF, has been observed (7). The value of rifampin for therapy of listeriosis remains a matter of debate. Whereas the in vitro activity is good (see above) there is only an additive or even an antagonistic effect when it is used in combination with ampicillin. This effect has been further substantiated by in vivo experiments on rabbit meningitis (113). On the other hand, in the mouse, where listeriae reside intracellularly, rifampin has a marked therapeutic effect (51). Thus, one could argue that for the initial therapy of listeriosis, which most often manifests itself as meningitis, rifampin is not essential, but for a complete cure, which would include the eradication of intracellular bacteria hidden, for example, within parenchymal cells of the brain, the use of rifampin can be assumed to be important. However, this assumption remains speculative at present (74).

Some recommendations for empiric therapy of bacterial meningitis include the use of cephalosporins (39). Those antibiotic regimens will be effective with most bacterial pathogens but not with *L. monocytogenes*, which require a selective treatment. Thus, an exact diagnosis is a prerequisite for a favorable outcome. In Gram-stained smears, *L. monocytogenes* can easily be mistaken for streptococci, so that an effective treatment is delayed. In listeria encephalitis, bacteria may reside entrapped in the infective foci and are not be reached by spinal puncture. As a result, diagnosis can not be made by culture, and serologic procedures for listeriosis are unreliable (48). Consequently, the difficult diagnosis may contribute to the high fatality rate because the best choice of antibiotics is missed.

CONCLUSIONS

Listeriosis is not a major problem in medicine, but the high mortality rate of overt listeriosis in very young, very old, and immunocompromised patients represents a challenge for veterinarians, food microbiologists, medical microbiologists, and clinicians. Since food has come to be regarded as the major source of infection, considerably preventive efforts have been taken by decontamination of domestic cattle and food items (27). The therapeutic efficacy of conventional treatment is still unsatisfactory, emphasizing that listeriosis remains a medical problem, at least in part because of the habitat of the organisms within host cells. Biologists have elucidated the intracellular behavior of *L. monocytogenes* (102, 124, 131).

In the most common clinical setting of meningitis, listeriae behave more as extracellular than intracellular pathogens, and therefore antibiotics that exert a good activity in vitro should be considered for the initial therapy of listeriosis. Since intrisic resistance except to cephalosporins is uncommon and acquired resistance is extremely rare, many options are available. Unfortunately, only few agents are rapidly bactericidal for *L. monocytogenes.*

Furthermore, because meningitis and encephalitis are the prominent clinical manifestations of disease, drugs that easily cross the blood-CSF barrier and the blood-brain barrier must be chosen. Whereas antibiotics may have a good chance to act on listeriae in the more permeable inflamed meninges and the ventricles, the blood-brain barrier is more complex in normal situations and possibly more restrictive even in the case of inflammation. Thus, in practice, the choice of therapy is limited in spite of the large array of antibiotics active in vitro. Ampicillin or, even better, amoxicillin, combined with an aminoglycoside remains the primary choice. The aminoglycoside should be added because it enhances the bactericidal action of penicillins. For patients allergic to β-lactams, co-trimoxazole represents an alternative drug.

Drugs are able to interact with extracellular bacteria and reduce the bacterial load. In normal immunocompetent patients, the body's defense system will rid itself of the residual bacteria, so that the therapeutic efficiency of antibiotics is generally good. On the other hand, because *L. monocytogenes* resides and multiplies intracellularly, manifestations of listeriosis other than meningitis, for example encephalitis, may develop. Listeriae can infect several types of brain cells, namely, ependymal cells, macrophages, microglial cells, and neurons (117). Neurons normally do not express major histocompatibility complex class I antigens (87), so that T-cell-mediated immunity against *L. monocytogenes* cannot rid these niches of the organism. Furthermore, no information about antibiotic accumulation in neurons is available. Hence, a high mortality rate in spite of administration of the recommended antibiotic regimen occurs and recurrence after initial improvement is often seen as well.

More from such theoretical considerations than from clinical experience, one may conclude that an optimal therapy of listeriosis may be achieved only when drugs that counteract intracellular bacteria are added. Rifampin is such a drug, but it may interfere with the action of ampicillin on extracellular listeriae (140).

Since listeriosis often occurs in cancer patients, one may assume that not only macrophages and parenchymal cells but also tumor cells will be infected by virulent *L. monocytogenes*, which can infect virtually every type of host cell (102, 124). A particular portion of such transformed cells will genetically express resistance to various cytostatic drugs (30, 32), including antibiotics. In addition, normal tissue cells may be induced to express MDR or other export systems. Hence, *L. monocytogenes* may persist in protected intracellular niches and infection may exacerbate after cessation of therapy. Thus, therapeutic failure may be due to resistance of host cells rather than to resistance of bacteria.

In the future, nonconventional methods will need to be applied to reduce the health risk posed by listeriae. The pos-

sibilities to prevent disease either by inhibition of *L. monocytogenes* in food or by interference with colonization of the gut, using bacteriocins, for example, should be considered further. Another possibility is the stimulation of innate, nonspecific defense mechanisms. Since *L. monocytogenes* is highly susceptible to endogenous antibiotics, such as defensins, the application or induction of such protective substances would be highly beneficial. Progress in the modulation of the specific immune reaction could also have an important impact on the treatment of listeriosis.

REFERENCES

1. **Acar, J., and F. W. Goldstein.** 1993. In vitro activity against gram-positive and gram-negative bacteria, p. 14–23. *In* H. Neu, L. S. Young, and S. H. Zinner (ed.), The new macrolides, azalides, and streptogramins. Marcel Dekker, Inc., New York, N.Y.
2. **Anonymous.** 1991. *Listeria monocytogenes*. Recommendations by the National Advisory Committee on microbiological criteria for foods. Int. J. Food Microbiol. **14:**185–246.
3. **Aseni, V., and J. Fierer.** 1991. Synergistic effect of human lysozyme plus ampicillin or β-lysin on the killing of *Listeria monocytogenes*. J. Infect. Dis. **163:**574–578.
4. **Azimi, P. H., K. Koranyi, and K. D. Lindsey.** 1979. *Listeria monocytogenes*. Synergistic effects of ampicillin and gentamicin. Am. J. Clin. Pathol. **72:** 974–977.
5. **Bakker-Woudenberg, I. A. J. M., A. F. Lokerse, F. H. Roerdink, D. Regts, and M. F. Michel.** 1985. Free versus liposome-entrapped ampicillin in treatment of infection due to *Listeria monocytogenes* in normal and athymic (nude) mice. J. Infect. Dis. **151:**917–924.
6. **Bakker-Woudenberg, I. A. J. M., A. F. Lokerse, J. C. Vink-van der Berg, and F. H. Roerdink.** 1988. Liposome-encapsulated ampicillin against *Listeria monocytogenes* in vivo and in vitro. Infection **16**(Suppl. 2)**:**165–170.
7. **Baldassarre, J. S., M. J. Ingerman, J. Nansteel, and J. Santoro.** 1991. Development of *Listeria* meningitis during vancomycin therapy: a case report. J. Infect. Dis. **164:**221–222.
8. **Bauer, J., H. Hof, T. Nichterlein, and M. Wuenscher.** 1993. Comparative activities of macrolide derivatives on murine listeriosis. Zentralbl. Bakteriol. **278:**112–119.
9. **Becq-Giraudon, B., and J. P. Breux.** 1987. Traitement des listérioses neuroméningées du sujet âgé de plus de soixante ans par l'association ampicilline + sulfaméthoxazole triméthoprime. Pathol. Biol. **35:**626–628.
10. **Beerens, H., and M. M. Tahon-Castel.** 1966. Milieu à l'acide nalidixique pour l'isolement des streptocoques, D. pneumoniae, Listeria, Erysipelothrix. Ann. Inst. Pasteur (Paris) **111:**90–93.
11. **Berner, R., and H. Hof.** 1988. Therapeutic activity of teicoplanin on experimental listeriosis compared with that of vancomycin and ampicillin. Zentralbl. Bakteriol. Mikrobiol. Hyg. Ser. A **268:**50–56.
12. **Blatt, S. P., and R. A. Zajac.** 1991. Treatment of listeria bacteremia with vancomycin. Rev. Infect. Dis. **13:**181–182.
13. **Boisivon, A., C. Guiomar, and C. Carbon.** 1990. In-vitro bactericidal activity of amoxicillin, gentamicin, rifampicin, ciprofloxacin and trimethoprim-sulfamethoxazole alone or in combination against *Listeria monocytogenes*. Eur. J. Clin. Microbiol. Infect. Dis. **9:**206–209.
14. **Cao, C. X., S. C. Silverstein, H. C. Neu, and T. H. Steinberg.** 1992. J774 macrophages secrete antibiotics via organic anion transporters. J. Infect. Dis. **165:**322–328.
15. **Charpentier, E., G. Gerbaud, C. Jaquet, J. Rocourt, and P. Courvalin.** 1995. Incidence of antibiotic resistance in Listeria species. J. Infect. Dis. **172:**277–281.
16. **Cherubin, C. E., M. D. Appleman, P. N. R. Heseltine, W. Khayr, and C. W. Stratton.** 1991. Epidemiological spectrum and current treatment of listeriosis. Rev. Infect. Dis. **13:**1108–1114.
17. **Doucet-Populaire, F., P. Trieu-Cuot, I. Dosbaa, A. Andremont, and P. Courvalin.** 1991. Inducible transfer of conjugative transposon Tn*1545* from *Enterococcus faecalis* to *Listeria monocytogenes* in the digestive tracts of gnotobiotic mice. Antimicrob. Agents Chemother. **35:**185–187.
18. **Drevets, D. A., B. P. Canono, P. J. M. Leenen, and P. A. Campbell.** 1994. Gentamicin kills intracellular *Listeria monocytogenes*. Infect. Immun. **62:** 2222–2228.
19. **Dryden, M. S., N. F. Jones, and I. Phillips.** 1991. Vancomycin therapy failure in *Listeria monocytogenes* peritonitis in a patient on continuous ambulatory peritoneal dialysis. J. Infect. Dis. **164:**1239.
20. **Edmiston, C. E., and R. C. Gordon.** 1979. Evaluation of gentamicin and penicillin as a synergistic combination in experimental murine listeriosis. Antimicrob. Agents Chemother. **16:**862–863.
21. **Ehlert, F., and H. C. Neu.** 1987. In-vitro activity of LY146032 (daptomycin), a new peptolide. Eur. J. Clin. Microbiol. **6:**84–90.
22. **Eliopoulos, G. M., and R. C. Moellering.** 1981. Susceptibility of enterococci and *Listeria monocytogenes* to *N*-formimidoyl thienamycin alone and in combination with an aminoglycoside. Antimicrob. Agents Chemother. **19:** 789–793.
23. **Elsbach, P., and J. Weiss.** 1988. Phagocytic cells: oxygen-independent antimicrobial systems, p. 445–470. *In* J. L. Gallin, I. M. Goldstein, and R. Snyderman (ed.), Inflammation: basic principles and clinical correlates. Raven Press, New York, N.Y.
24. **Espaze, E. P., and A. E. Reynaud.** 1988. Antibiotic susceptibilities of *Listeria*: in vitro studies. Infection **16**(Suppl. 2)**:**160–164.
25. **Facinelli, B., P. E. Giovanetti, Varaldo, P., Casolari, P., and U. Fabio.** 1991. Antibiotic resistance in foodborne listeria. Lancet **338:**1272.
26. **Fiedler, F.** 1988. Biochemistry of the cell surface of *Listeria* strains: a locating general view. Infection **16**(Suppl. 2)**:**92–97.
27. **Gellin, B. G., and C. V. Broome.** 1989. Listeriosis. JAMA **261:**1313–1318.
28. **Gentschev, I., Z. Sokolovic, S. Köhler, G. F. Krohne, H. Hof, J. Wagner, and W. Goebel.** 1992. Identification of p60 antibodies in human sera and presentation of this listerial antigen on the surface of attenuated salmonellae by the hlyB-hlyD secretion system. Infect. Immun. **60:**5091–5098.
29. **Goetz, F. (University of Tübingen, Tübingen, Germany).** Personal communication.
30. **Goldstein, L. J., H. Galski, A. T. Fojo, M. C. Willingham, S. J. Lai, A. Gazdar, R. Pirker, A. Green, W. Crist, G. Rodeur, C. Grant, M. Lieber, J. Cossman, M. M. Gottesman, and I. Pastan.** 1989. Expression of a multidrug resistance gene in human cancers. J. Natl. Cancer Inst. **81:**116–124.
31. **Gordon, R. C., R. E. Wofford, T. Hsu, and L. L. Fechner.** 1980. Ampicillin-gentamicin synergism against *Listeria monocytogenes* in vitro. Infection **8:**221–222.
32. **Gottesman, M. M., and I. Pastan.** 1993. Biochemistry of multidrug resistance mediated by the multidrug transporter. Annu. Rev. Biochem. **62:**385–427.
33. **Groisman, E. A.** 1994. How bacteria resist killing by host-defense peptides. Trends Microbiol. **2:**444–449.
34. **Gückl, H., and H. Hof.** 1986. Vergleich der therapeutischen Wirkung von verschiedenen Penicillinderivaten und Imipenem bei der experimentellen Listeriose. Z. Antimikrob. Antineoplast. Chemother. **4:**84–87.
35. **Günther, G., and A. Philipson.** 1988. Oral trimethoprim as follow-up treatment of meningitis caused by Listeria monocytogenes. Rev. Infect. Dis. **10:**53–55.
36. **Hadorn, K., H. Hächler, H., A. Schaffner, and F. H. Kayser.** 1993. Genetic characterization of plasmid-encoded multiple antibiotic resistance in a strain of *Listeria monocytogenes* causing endocarditis. Eur. J. Clin. Microbiol. Infect. Dis. **12:**928–937.
37. **Hakenbeck, R., and H. Hof.** 1991. Relatedness of penicillin-binding proteins from various Listeria species. FEMS Microbiol. Lett. **84:**191–196.
38. **Héchard, Y., C. Jayat, F. Letellier, M. H. Ratinaud, R. Julien, and Y. Cenatiempo.** 1992. A new Leuconostoc bacteriocin, mesentericidin Y105, bacteriocidal to *Listeria monocytogenes*. NATO ASI Ser. Ser. H **65:**421–446.
39. **Helwig, H.** 1984. Empfehlungen der Arbeitsgemeinschaft "Meningitis" der Paul-Ehrlich-Gesellschaft für Chemotherapie e. V. zur Diagnostik und Therapie der bakteriellen Meningitis. Z. Antimikrob. Antineoplast. Chemother. **2:**209–213.
40. **Heymer, B., C. H. Wirsing von König, H. Finger, H. Hof, and P. Emmerling.** 1988. Histomorphology of experimental listeriosis. Infection **16**(Suppl. 2)**:** 106–111.
41. **Hiemstra, P. S., P. B. Eisenhauer, S. S. L. Harwig, M. T. van den Barselaar, R. Furth van, and I. R. Lehrer.** 1993. Antimicrobial proteins of murine macrophages. Infect. Immun. **61:**3038–3046.
42. **Hodinka, R. L., K. Jack-Wait, N. Wannamaker, T. P. Walden, and P. H. Gilligan.** 1987. Comparitive in vitro activity of Ly146032 (daptomycin), a new lipopeptide antimicrobial. Eur. J. Clin. Microbiol. **6:**100–103.
43. **Hof, H.** 1984. Virulence of different strains of *Listeria monocytogenes* serovar 1/2a. Med. Microbiol. Immunol. **173:**207–218.
44. **Hof, H.** 1986. The effect of coumermycin on experimental listeriosis. J. Infect. **13:**17–23.
45. **Hof, H.** 1987. Failure of recombinant murine τ-interferon to cure chronic listeriosis of nude, athymic mice. Thymus **10:**247–249.
46. **Hof, H.** 1989. Die Wirkung von Daptomycin (LY146032) auf Listerien in vitro und in vivo. Z. Antimikrob. Antineoplast. Chemother. **7:**35–38.
47. **Hof, H.** 1990. Treatment of experimental listeriosis by CI 934, a new quinolone. J. Antimicrob. Chemother. **25:**121–126.
48. **Hof, H.** 1990. Listerien—eine Herausforderung für die Diagnostik. Immun. Infekt. **18:**35–39.
49. **Hof, H.** 1991. Intracellular microorganisms: a particular problem for chemotherapy. Infection **19**(Suppl. 4)**:**193–194.
50. **Hof, H.** 1991. Therapeutic activities of antibiotics in listeriosis. Infection **19**(Suppl. 4)**:**229–233.
51. **Hof, H., and P. Emmerling.** 1984. Murine model for therapy of listeriosis in the compromised host. III. The effect of rifampicin. Chemotherapy **30:**125–130.
52. **Hof, H., and J. Fabrig.** 1987. Die Wirkung von Gyrase-Hemmern auf Listerien. Fortschr. Antimikrob. Antineoplast. Chemother. **6-10:**1781–1785.
53. **Hof, H., and H. Gückl.** 1987. Lack of synergism of ampicillin and gentamicin in experimental listeriosis. Infection **15:**40–41.

54. **Hof, H., and G. Waldenmeier.** 1988. Therapy of experimental listeriosis—an evaluation of different antibiotics. Infection **16**(Suppl.)**2:**171–174.
55. **Jack, R. W., J. R. Tagg, and B. Ray.** 1995. Bacteriocins of gram-positive bacteria. Microbiol. Rev. **59:**171–200.
56. **James, R., C. Lazdunski, and H. Pattus (ed.).** 1992. Plasmid coded toxins: bacteriocins, microcins and lantibiotics. Springer-Verlag KG, Heidelberg, Germany.
57. **Jones, E. M., and A. P. MacGowan.** 1995. Antimicrobial chemotherapy of human infection due to *Listeria monocytogenes*. Eur. J. Clin. Microbiol. Infect. Dis. **14:**165–175.
58. **Kalstone, C.** 1991. Succesful antepartum treatment of listeriosis. Am. J. Obstet. Gynecol. **164:**57–58.
59. **Kiderlen, A. F., S. H. E. Kaufmann, and M. L. Lohmann-Matthes.** 1984. Protection of mice against the intracellular bacterium *Listeria monocytogenes* by recombinant immune interferon. Eur. J. Immunol. **14:**964–967.
60. **Kihlström, E.** 1977. Infection of HeLa cells with *Salmonella typhimurium*. Infect. Immun. **17:**290–295.
61. **Kim, K. S.** 1986. In vitro and in vivo studies of imipenem-cilastatin alone and in combination with gentamicin against *Listeria monocytogenes*. Antimicrob. Agents Chemother. **29:**289–293.
62. **Kishore, B. K., P. Maldague, and P. M. Tulkens.** 1992. Lysosomal storage disorders and toxicity induced by aminoglycoside antibiotics and polyanionic peptides, p. 439–445. *In* P. J. Courtoy (ed.), Endocytosis. Springer-Verlag KG, Berlin, Germany.
63. **Kretschmar, M., C. Budeanu, T. Nichterlein, and H. Hof.** 1993. Wirkung von Mersacidin auf die murine Listeriose. Chemother. J. **2:**176–178.
64. **Lavetter, A., J. L. Leedom, A. W. Mathies, D. Ivler, and P. F. Wehrle.** 1971. Meningitis due to *Listeria monocytogenes*. A review of 25 cases. N. Engl. J. Med. **285:**598–603.
65. **Lehrer, R. I.** 1993. Defensins: antimicrobial and cytotoxic peptides of mammalian cells. Annu. Rev. Immunol. **11:**105–128.
66. **Lorber, B., J. Santoro, and R. M. Seenson.** 1975. Listeria meningitis during cefazolin therapy. Ann. Intern. Med. **82:**226.
67. **Louria, D. B., T. Hensle, D. Armstrong, H. S. Collins, A. Blevins, D. Krugman, and M. Buse.** 1967. Listeriosis complicating malignant disease: a new association. Ann. Intern. Med. **67:**261–281.
68. **Loza, E., J. Martinez Beltran, F. Baquero., A. Léon, R. Cantón, and B. Garijo.** 1992. Comparative in vitro activity of clarithromycin. Eur. J. Clin. Microbiol. Infect. Dis. **11:**856–866.
69. **MacGowan, A. P., H. A. Holt, and D. S. Reeves.** 1990. In vitro synergy testing of nine antimicrobial combinations against *Listeria monocytogenes*. J. Antimicrob. Chemother. **25:**561–566.
70. **MacGowan, A. P., H. A. Holt, M. J. Bywater, and D. S. Reeves.** 1990. In vitro antimicrobial susceptibility of *Listeria monocytogenes* isolated in the UK and other *Listeria* species. Eur. J. Clin. Microbiol. Infect. Dis. **9:**767–770.
71. **Mackaness, G. B.** 1962. Cellular resistance to infection. J. Exp. Med. **116:** 381–406.
72. **Macnair, D. R., J. E. White, and J. M. Graham.** 1968. Ampicillin in the treatment of *Listeria monocytogenes* meningitis. Lancet **i:**16–17.
73. **Mangi, R. J., R. S. Kundargi, R. Quintiliani, and V. T. Andriole.** 1973. Development of meningitis during cephalothin therapy. Ann. Intern. Med. **78:**347–351.
74. **Marget, W., and H. P. R. Seeliger.** 1988. *Listeria monocytogenes* infections—therapeutic possibilities and problems. Infection **16**(Suppl. 2)**:**175–177.
75. **Marklein, G., and J. Häfelein.** 1981. Zur Empfindlichkeit in vitro von *Listeria monocytogenes* unter besonderer Berücksichtigung neuerer Beta-Laktamantibiotika. Zentralbl. Bakteriol. Mikrobiol. Hyg. Abt. 1 Orig. A **251:**40–53.
76. **Marquardt, D., S. McCrone, and M. S. Center.** 1990. Mechanisms of multidrug resistance in HL 60 cells: detection of resistance-associated proteins with antibodies against synthetic peptides that correspond to the deduced sequence of P-glycoprotein. Cancer Res. **50:**1426–1430.
77. **McCracken, G. H., J. D. Nelson, and M. L. Thomas.** 1973. Discrepancy between carbenicillin and ampicillin activities against enterococci and *Listeria* spp. Antimicrob. Agents Chemother. **3:**343–349.
78. **McLauchlin, J., A. Audurier, and A. G. Taylor.** 1991. Treatment failure and recurrent human listeriosis. J. Antimicrob. Chemother. **27:**851–857.
79. **Medoff, G., L. J. Kunz, and A. N. Weinberg.** 1971. Listeriosis in humans. An evaluation. J. Infect. Dis. **123:**247–250.
80. **Metz, P., H. Hof, and S. Kathariou.** 1988. Inhibitory effect of tetracycline and doxycycline on resistance of mice to infection with a tetracycline-resistant strain of *Listeria monocytogenes*. Int. J. Immunopharmacol. **10:** 519–524.
81. **Michelet, C., J. L. Avril, F. Cartier, and P. Berche.** 1994. Inhibition of intracellular growth of *Listeria monocytogenes* by antibiotics. Antimicrob. Agents Chemother. **38:**438–446.
82. **Moellering, R. C., G. Medoff, I. Leech, C. Wennersten, and L. J. Kunz.** 1972. Antibiotic synergism against *Listeria monocytogenes*. Antimicrob. Agents Chemother. **1:**30–34.
83. **Mohan, K., R. C. Gordon, T. C. Beaman, R. C. Belding, D. Luecke, C. Edmiston, and P. Gerhardt.** 1977. Synergism of penicillin and gentamicin against *Listeria monocytogenes* in ex vivo hemodialysis culture. J. Infect. Dis. **135:**51–54.
84. **Nelson, J. D., S. Shelton, and D. Parks.** 1967. Antibiotic susceptibility of *Listeria monocytogenes* and treatment of neonatal listeriosis with ampicillin. Acta Paediatr. Scand. **56:**151–158.
85. **Nettles, C. G., and S. F. Barefoot.** 1993. Biochemical and genetic characteristics of bacteriocins of food-associated lactic acid bacteria. J. Food Prot. **56:**338–356.
86. **Neu, H., and P. Labthavikul.** 1983. In vitro activity of teichomycin compared with those of other antibiotics. Antimicrob. Agents Chemother. **24:** 425–428.
87. **Neumann, H., A. Cavalié, D. Jeune, and H. Wekerle.** 1995. Induction of MHC class I genes in neurons. Science **269:**549–552.
88. **Nichterlein, T., J. Bauer, and H. Hof.** 1992. Clarithromycin is superior to erythromycin: a tissue culture model of infection with *Listeria monocytogenes*. Med. Microbiol. Lett. **1:**85–90.
89. **Nichterlein, T., and H. Hof.** 1991. Effect of various antibiotics on *Listeria monocytogenes* multiplying in L929 cells. Infection **19**(Suppl. 4)**:**234–238.
90. **Nichterlein, T., M. Kretschmar, C. Budeanu, J. Bauer, W. Linss, and H. Hof.** 1994. Bay Y 3118, a new quinolone derivative, rapidly eradicates *Listeria monocytogenes* from infected mice and L929 cells. Antimicrob. Agents Chemother. **38:**1501–1506.
91. **Nichterlein, T., M. Kretschmar, and H. Hof.** 1996. RP 59500, a streptogramin derivative, is effective in murine listeriosis. J. Chemother. **8:**107–112.
92. **Nichterlein, T., M. Kretschmar, M. Siegsmund, and H. Hof.** 1995. Erythromycin is ineffective against *Listeria monocytogenes* in multidrug resistant cells. J. Chemother. **7:**184–188.
93. **Nichterlein, T., E. Domann, M. Kretschmar, M. Bauer, A. Hlawatsch, H. Hof, and T. Chakraborty.** 1996. Subinhibitory concentrations of β-lactams and other cell-wall antibiotics inhibit listeriolysin production by *Listeria monocytogenes*. Int. J. Antimicrob. Agents **7:**75–81.
94. **Niemann, R. E., and B. Lorber.** 1980. Listeriosis in adults: a changing pattern. Report of 8 cases and review of the literature. Rev. Infect. Dis. **2:**207–227.
95. **Odio, C., M. L. Thomas, and G. H. McCracken.** 1984. Pharmacokinetics and bacteriological efficacy of mezlocillin in experimental *Escherichia coli* and *Listeria* meningitis. Antimicrob. Agents Chemother. **25:**427–432.
96. **Parente, E., and C. Hill.** 1992. Inhibition of *Listeria* in buffer, broth, and milk by enterocin 1146, a bacteriocin produced by *Enterococcus faecium*. J. Food Prot. **55:**503–508.
97. **Payne, J., and M. Gasson.** 1992. Cloning and characterization of a lysin gene from *Listeria* bacteriophage. NATO ASI Ser. Ser. H **65:**427–431.
98. **Penn, R. L., T. T. Ward, and R. T. Steigbiegel.** 1982. Effects of erythromycin in combination with penicillin, ampicillin, or gentamicin on the growth of *Listeria monocytogenes*. Antimicrob. Agents Chemother. **22:**289–294.
99. **Pierre, J., A. Boisivon, and L. Gutmann.** 1990. Alteration of PBP3 entails resistance to imipenem in *Listeria monocytogenes*. Antimicrob. Agents Chemother. **34:**1695–1698.
100. **Pollock, S. S., T. M. Pollock, and M. J. G. Harrison.** 1986. Ampicillin-resistant *Listeria monocytogenes* meningitis. Arch. Neurol. **43:**106.
101. **Porter, J. R., and W. H. Hale.** 1939. Effect of sulfanilamide and sulfapyridine on experimental infections with Listerella and Erysipelothrix in mice. Proc. Soc. Exp. Biol. Med. **42:**47–50.
102. **Portnoy, D. A., T. Chakraborty, W. Goebel, and P. Cossart.** 1992. Molecular determinants of *Listeria monocytogenes* pathogenesis. Infect. Immun. **60:** 1263–1270.
103. **Poyart-Salmeron, C., C. Carlier, P. Trieu-Cuot, A. L. Courtieu, and P. Courvalin.** 1990. Transferable plasmid-mediated antibiotic resistance in *Listeria monocytogenes*. Lancet **335:**1422–1426.
104. **Poyart-Salmeron, C., P. Trieu-Cuot, C. Carlier, A. MacGowan, J. McLauchlin, and P. Courvalin.** 1992. Genetic basis of tetracycline resistance in clinical isolates of *Listeria monocytogenes*. Antimicrob. Agents Chemother. **36:**463–466.
105. **Pucci, M. J., E. R. Vedamuthu, B. S. Kunka, and P. A. Vandenbergh.** 1988. Inhibition of *Listeria monocytogenes* by using bacteriocin PA-1 produced by *Pediococcus acidilactici* PAC 1.0. Appl. Environ. Microbiol. **54:**2349–2353.
106. **Quentin, C., M. C. Thibaut, J. Horovits, and C. Bebear.** 1990. Multiresistant strain of *Listeria monocytogenes* in septic abortion. Lancet **336:**375.
107. **Raoult, D., M. Drancourt, and G. Vestris.** 1990. Bactericidal effect of doxycycline associated with lysosomotropic agents on *Coxiella burnetii* in P388 D_1 cells. Antimicrob. Agents Chemother. **34:**1512–1514.
108. **Rapp, M. F., H. A. Pershadsingh, J. W. Long, and J. M. Pickens.** 1984. Ampicillin-resistant *Listeria monocytogenes* meningitis in a previously healthy 14-year-old athlete. Arch. Neurol. **41:**1304.
109. **Reynaud, H., E. P. Espaze, S. Papin, and A. L. Courtieu.** 1984. Etude de la sensibilité aux antibiotiques de 139 souches de *Listeria* serotypées par le Centre National de Réference en 1983. Ann. Microbiol. (Paris) **135B:**331–339.
110. **Sanchez, M. L., R. P. Wenzel, and R. N. Jones.** 1992. In vitro activity of decaplanin (M86-1410), a new glycopeptide antibiotic. Antimicrob. Agents Chemother. **36:**873–875.

111. **Sande, M. A.** 1981. Factors influencing the penetration and activity of antibiotics in experimental meningitis. J. Infect. **3**(Suppl. 1)**:**33–38.
112. **Satta, G., G. Cornaglia, A. Mazzariol, G. Golini, S. Valisena, and R. Fontana.** 1995. Target for bacteriostatic and bactericidal activities of β-lactam antibiotics against *Escherichia coli* resides in different penicillin-binding proteins. Antimicrob. Agents Chemother. **39:**812–818.
113. **Scheld, W. M.** 1983. Evaluation of rifampicin and other antibiotics against *Listeria monocytogenes* in vitro and in vivo. Rev. Infect. Dis. **5**(Suppl. 3)**:** 593–599.
114. **Scheld, W. M., D. D. Fletcher, F. N. Fink, and M. A. Sande.** 1979. Response to therapy in an experimental rabbit model of meningitis due to *Listeria monocytogenes*. J. Infect. Dis. **140:**287–294.
115. **Schillinger, U., B. Becker, and W. H. Holzapfel.** 1995. Antibacterial activity of carnocin 54, a bacteriocin from *Leuconostoc carnosum*. Food Microbiol. **12:**31–37.
116. **Schillinger, U., M. Kaya, and F. K. Lücke.** 1991. Behaviour of *Listeria monocytogenes* in meat and its control by a bacteriocin-producing strain of *Lactobacillus sake*. J. Appl. Bacteriol. **70:**473–478.
117. **Schlüter, D., S. Chahoud, H. Lassmann, A. Schumann, H. Hof, and M. Deckert-Schlüter.** 1996. Intracerebral targets and immunomodulation of murine *Listeria monocytogenes* meningoencephalitis. J. Neuropathol. Exp. Neurol. **55:**14–24.
118. **Schmitt-Slomska, J., M. C. Marmouset, C. Louis, R. Bally, G. Starka, and S. Madoff.** 1982. Tetracycline-resistant L-forms isolated from an antibiotic-susceptible strain of *Listeria monocytogenes*. Antimicrob. Agents Chemother. **22:**678–685.
119. **Schröter, G., and R. Oppel.** 1983. *Listeria monocytogenes*—in vitro Aktivität von Azlocillin und 13 weiteren Antibiotika. Muench. Med. Wochenschr. **125**(Suppl. 2)**:**159–162.
120. **Schwarzkopf, A.** 1986. Untersuchungen zur Bakterizidie von Ampicillin und Amoxycillin gegenüber *Listeria monocytogenes*. Doctoral thesis, University of Würzburg, Würzburg, Germany.
121. **Seeliger, H. P. R., U. Laymann, and H. Finger.** 1967. Untersuchungen zur Frage der Wirksamkeit von Ampicillin auf Listeria monocytogenes. Dtsch. Med. Wochenschr. **92:**1095–1097.
122. **Selsted, M. E., D. Szklarek, and R. I. Lehrer.** 1984. Purification and antibacterial activity of antimicrobial peptides of rabbit granulocytes. Infect. Immun. **45:**150–154.
123. **Seoh, S. A., and D. Busath.** 1993. The permeation properties of small organic cations in gramicidin A channels. Biophys. J. **64:**1017–1028.
124. **Sheehan, B., C. Kocks, S. Dramsi, E. Gouin, A. D. Klarsfeld, J. Mengaud, and P. Cossart.** 1994. Molecular and genetic determinants of the *Listeria monocytogenes* infectious process. Curr. Top. Microbiol. Immunol. **192:** 187–216.
125. **Siragusa, G. R., and C. Nettles Cutter.** 1993. Brochocin-C, a new bacteriocin produced by *Brochotrix campestris*. Appl. Environ. Microbiol. **59:**2326–2328.
126. **Skogberg, K., J. Syrjänen, M. Jahkola, O. V. Renkonen, J. Paavonen, S. Kontiainen, P. Ranta, and V. Valtonen.** 1992. Clinical presentation and outcome of listeriosis in patients with and without immunosuppressive therapy. Clin. Infect. Dis. **14:**815–823.
127. **Slade, P. J., and D. L. Collins-Thompson.** 1990. Listeria, plasmids, antibiotic resistance, and food. Lancet **i:**1004.
128. **Spitzer, P. G., S. M. Hammer, and A. W. Karchmer.** 1986. Treatment of *Listeria monocytogenes* infection with trimethoprim-sulfamethoxazole: case report and review of the literature. Rev. Infect. Dis. **8:**427–430.
129. **Squinazi, F., J. Bourgade, and P. Geslin.** 1986. Activités bactériostatiques et bactericides des antibiotiques sur *Listeria monocytogenes*, p. 456–459. *In* A. L. Courtieu, E. P. Espaze, and A. E. Reynaud (ed.), Listeriose, listeria, listeriosis. University of Nantes, Nantes, France.
130. **Testa, R. T., P. J. Petersen, N. V. Jacobus, P. E. Sum, V. J. Lee, and F. P. Tally.** 1993. In vitro and in vivo antibacterial activities of the glycylcyclines, a new class of semisynthetic tetracyclines. Antimicrob. Agents Chemother. **37:**2270–2277.
131. **Tilney, I. G., and M. S. Tilney.** 1993. The wily ways of a parasite: induction of actin assembly by Listeria. Trends Microbiol. **1:**25–31.
132. **Tsurno, T., T. Tsierno, H. Hamada, M. M. Gottesman, I. Pastan, and M. C. Willingham.** 1987. Cellular localization of the multidrug resistance gene product P-glycoprotein in normal human tissues. Proc. Natl. Acad. Sci. USA **84:**7735–7738.
133. **Tuazon, C. U., D. Shamsuddin, and H. Miller.** 1982. Antibiotic susceptibility and synergy of clinical isolates of *Listeria monocytogenes*. Antimicrob. Agents Chemother. **21:**525–527.
134. **van der Auwera, P., P. Grenier, and J. Klastersky.** 1987. In-vitro activity of LY146032 against *Staphylococcus aureus*, *Listeria monocytogenes*, *Corynebacterium* JK, and *Bacillus* spp., in comparison with various antibiotics. J. Antimicrob. Chemother. **20:**209–212.
135. **van Laack, R. L. J. M., U. Schillinger, and W. H. Holzapfel.** 1992. Characterization and partial purification of a bacteriocin produced by *Leuconostoc carnosum* LA44A. Int. J. Food Microbiol. **16:**184–195.
136. **Vicente, M. F., J. C. Perez-Diaz, F. Baquero, M. Angel de Pedro, and J. Berenguer.** 1990. Penicillin-binding protein 3 of *Listeria monocytogenes* as the primary lethal target for β-lactams. Antimicrob. Agents Chemother. **34:**539–542.
137. **Vischer, W. A., and C. Rominger.** 1978. Rifampicin against experimental listeriosis in the mouse. Chemotherapy **24:**104–111.
138. **Weingärtner, L., and S. Ortel.** 1967. Zur Behandlung der Listeriose mit Ampicillin. Dtsch. Med. Wochenschr. **92:**1098–1104.
139. **Wiggins, G. L., W. L. Albritton, and J. C. Feeley.** 1978. Antibiotic susceptibility of clinical isolates of *Listeria monocytogenes*. Antimicrob. Agents Chemother. **13:**854–860.
140. **Winslow, D. L., J. Damme, and E. Diekman.** 1983. Delayed bactericidal activity of β-lactam antibiotics against *Listeria monocytogenes*: antagonism of chloramphenicol and rifampin. Antimicrob. Agents Chemother. **23:**555–558.
141. **Winslow, D. L., and M. L. Steele.** 1984. Listeria bacteremia and peritonitis associated with a peritoneovenous shunt: successful treatment with sulfamethoxazole and trimethoprim. J. Infect. Dis. **149:**820.
142. **Winslow, D. L., and G. A. Pankey.** 1982. In vitro activities of trimethoprim and sulfamethoxazole against *Listeria monocytogenes*. Antimicrob. Agents Chemother. **22:**51–54.
143. **Wirsing von König, C. H., B. Heymer, H. Finger, P. Emmerling, and H. Hof.** 1988. Alteration of non-specific resistance to infection with *Listeria monocytogenes*. Infection **16**(Suppl. 2)**:**112–117.
144. **Yersin, B. R., M. P. Glauser, and F. Regli.** 1981. Infections à *Listeria monocytogenes* chez l'adulte. Schweiz. Med. Wochenschr. **111:**1596–1602.