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Sequence Homology of Clindamycin Resistance Determinants in Clinical Isolates of *Bacteroides* spp.

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The sequence homology of clindamycin resistance (Cln^r) determinants was studied in 16 Cln^r *Bacteroides* strains. The isolates were surveyed for plasmid content, homology with the Cln^r determinant of pBFTM10, and ability to transfer Cln^r. The Cln^r DNA probes used in the Southern hybridizations were pBFTM10 and a plasmid derivative containing an *Eco*RI fragment of pBFTM10 cloned into *Escherichia coli*. A total of 13 of 16 Cln^r strains also carried tetracycline resistance, and 15 of 16 Cln^r *Bacteroides* isolates showed homology with the Cln^r determinant of pBFTM10. These data suggest that the previously characterized Cln^r determinant of pBFTM10 is widely distributed in nature and may be found on either a plasmid or the chromosome. The Cln^r *Bacteroides fragilis* strain which lacked homology with pBFTM10 also had different transfer properties; thus, more than one type of Cln^r determinant may exist in *Bacteroides* spp.

The *Bacteroides fragilis* group, which includes *B. fragilis, Bacteroides thetaiotaomicron, Bacteroides vulgatus, Bacteroides ovatus*, and *Bacteroides distasonis*, contains strains which differ from most other anaerobic bacteria in that most are resistant to the β-lactam antibiotics and 60% are resistant to tetracycline (6). *B. fragilis* stands out as a dominant pathogen in the *B. fragilis* group because of its frequency of isolation in intraabdominal and gynecological pelvic infections and its tendency to cause bacteremia (10). Before the 1960s most strains of *B. fragilis* were susceptible to tetracycline.

Clindamycin, a lincosamide antibiotic, has become one of the primary agents for treatment of infections involving *Bacteroides* spp. because of its excellent in vitro activity, its efficacy in animal models, and the results of prospective controlled studies in the treatment of mixed anaerobic-aerobic infections (21). Clindamycin has maintained its position as a prime therapeutic agent in these clinical settings because relatively few resistant isolates have been documented (20). Although the occurrence of clindamycin-resistant (Cln^r) *Bacteroides* strains has been infrequent, there has been one reported outbreak of clindamycin resistance in Detroit, Mich., as well as scattered reports of resistant isolates (1, 13).

However, the potential for an increasing incidence of Cln^r strains exists. This view is based on several studies demonstrating plasmid-mediated conjugal transfer of clindamycin-erythromycin resistance in *Bacteroides* spp. and the widespread use of these agents (11, 18, 24). There are two known Cln^r transfer factors in *Bacteroides* spp.: pBF4 (pIP410), described by two groups (8, 25), and pBFTM10, which has been described by our group (19). Homology between the Cln^r determinants of pBF4 and pBFTM10 has been recently established (14). The present report examines the relatedness between 16 Cln^r strains of *Bacteroides* and the Cln^r determinant in pBFTM10.

MATERIALS AND METHODS

Bacteriology. The identification of *Bacteroides* isolates referred to our laboratory was confirmed by criteria (7) developed at the Virginia Polytechnic Institute and State University, Blacksburg. All identification tests, susceptibility studies, and transfer experiments were performed in an anaerobic chamber at 37°C (18). Strains of *B. fragilis* were classified according to a phage typing system developed in our laboratory (A. Sosa, M. H. Malamy, S. L. Gorbach, and F. P. Tally, unpublished data). Organisms were preserved in skim milk at −70°C. Bacteria were retrieved from frozen stocks and inoculated onto blood agar plates; single colonies were transferred to brain heart infusion broth–0.0005% hemin–0.5% yeast extract (BHIS) agar plates containing 10 μg of clindamycin per ml for use in the experiments described below.

Media. BHIS was used for liquid culture. The solid medium used for susceptibility testing was BHIS with 1.5% agar and 5% defibrinated sheep blood. The *B. fragilis* minimal medium used to confirm transconjugants was prepared as previously described (18). Antimicrobial susceptibility testing was performed by microtiter techniques adapted to the anaerobic chamber, using BHIS broth and an inoculum of approximately 10^6 CFU/ml as previously described (16). Sensitivity to virulent bacteriophage was determined by the agar

overlay method. BHIS agar plates with soft agar overlays were prepared under aerobic conditions and then incubated in the anaerobic chamber.

DNA analysis. Plasmid and chromosomal DNA were obtained by a Triton X-100 minilysis technique from 10-ml cultures as described previously (3) except for an incubation of the lysis mix at 65°C for 5 min. The DNA-containing supernatant was collected by centrifugation at 9,000 × *g* for 40 min. Plasmid DNA for nick translation was purified by harvesting the lower band in cesium chloride-ethidium bromide density gradient centrifugation of the cleared lysates (18). DNA samples were analyzed by electrophoresis in vertical agarose slab gels as previously described (18).

DNA homology studies. DNA hybridization studies were carried out by the technique of Southern (15). DNA was prepared by the minilysis technique described above. DNA was transferred to nitrocellulose paper and probed with ^{32}P-nick-translated DNA (12). The Cln^r transfer factor pBFTM10 or the *Eco*RI-B fragment of this plasmid cloned in *Escherichia coli* (9; M. H. Malamy, M. J. Shimell, and F. P. Tally, unpublished data) was used as a probe. The rationale for the use of these probes is discussed below.

Resistance transfer experiments and mating controls. The transferability of Cln^r in the *Bacteroides* strains was tested by utilizing the filter mating technique (18). The recipient strain was *B. fragilis* JC101, a plasmid-free auxotroph of strain TM4000 (see Table 1), which requires histidine and arginine for growth in minimal media. Strain JC101 is clindamycin susceptible (Cln^s), tetracycline susceptible (Tet^s), rifampicin resistant (Rif^r), and sensitive to phage FP (M. H. Malamy, G. R. Carson, and F. P. Tally, unpublished data). All strains used as donors were Cln^r Tet^r Rif^s. Transconjugants were selected on BHIS plates containing 10 μg of clindamycin and 50 μg of rifampicin per ml. JC101 transconjugants were verified by their lack of growth on minimal medium, growth on minimal medium containing histidine and arginine, and sensitivity to phage FP. For studies on the induction of transfer proficiency, donor cells were grown in a low concentration (1 μg/ml) of tetracycline or clindamycin before the donor and recipient were combined on the filter. In all matings, a TMP10 × JC101 cross was included as a positive control (see Table 1).

RESULTS

Source of Cln^r bacterial isolates. The source and selected characteristics of 16 Cln^r strains of *Bacteroides* and 3 control strains of *B. fragilis* are listed in Table 1. Ten strains were obtained from Betty Bond of the University of Illinois Clinical Microbiology Laboratory. All were highly resistant to clindamycin, with minimal inhibitory concentrations of clindamycin ranging from 128 to 512 μg/ml. In addition, they were all resistant to tetracycline (minimal inhibitory concentration, ≥8 μg/ml). When tested with a series of virulent bacteriophages that have been used to classify *B. fragilis*, each of the Cln^r *B. fragilis* isolates exhibited a different phage sensitivity pattern. This result implies that the different isolates were not closely related and did not represent repeated isolation of a single Cln^r strain. The six other Cln^r strains were obtained from various locations (Table 1). These strains had clindamycin minimal inhibitory concentrations of 256 to 512 μg/ml. Three of these strains were Tet^r, and three (TAL1877, TAL1912, and TAL1913) were Tet^s. These *B. fragilis* isolates could be assigned to three different phage sensitivity groups.

DNA analysis. A DNA analysis of the 16 isolates revealed that 10 strains were apparently plasmid free, whereas 6 isolates contained two to three bands of plasmid DNA after agarose gel electrophoresis (Table 1). The molecular sizes of the plasmids range from 2.8 to approximately 90 kilobases. The presence of very large plasmids (greater than 100 kilobases) in the "plasmid-free" strains cannot be ruled out at this time. These plasmids might not have been detected by the lysis technique used in this study.

Rationale for DNA homology studies. The Cln^r transfer factor pBFTM10 is useful as a probe in hybridization studies. Preliminary studies in our laboratory with strain TAL2300 revealed that ^{32}P-labeled pBFTM10 DNA has homology with the chromosome of this strain (17). This probe was subsequently used to study clindamycin resistance in Cln^r *Bacteroides* strains referred to our laboratory. Because of the low yields of DNA from *B. fragilis* and the low efficiency of incorporation of [^{32}P]triphosphate into *B. fragilis* DNA, the three *Eco*RI fragments of pBFTM10 were ligated into the *E. coli* plasmid vector pMC1403 and grown in *E. coli* (2, 9; Malamy, Shimell and Tally, unpublished data). Thus, large amounts of *B. fragilis* DNA could be harvested from *E. coli*. There was no homology between the vector pMC1403 and pBFTM10 (9). The 4.4-kilobase *Eco*RI fragment of pBFTM10 (*Eco*RI-B) is thought to contain the Cln^r determinant, based on the observation that loss of this segment of DNA results in a loss of clindamycin resistance (19). In addition, only this fragment shows homology with the Cln^r transfer factor pBF4 (14). The plasmid containing this fragment cloned in *E. coli*, designated pMJS100, was used for subsequent DNA homology studies.

DNA homology. Two different sources of Cln^r determinant DNA were used to test for the presence of related sequences in the Cln^r *Bacteroides* isolates. Plasmid pBFTM10 was used initially as a probe, and homology was revealed in 11 of the 16 isolates tested. The five isolates failing to hybridize with pBFTM10 were then tested with ^{32}P-labeled pMJS100. This DNA could be labeled to a higher specific activity by nick translation and was a more sensitive probe. Only strain TAL2450 failed to show homology with this probe; thus, 15 of 16 strains tested

TABLE 1. Bacteroides strains

TAL[a] no.	Species	Origin	MIC[b] (μg/ml)		Plasmids detected[c]	Homology with intact pBFTM10 or the cloned Cln[r] determinant pMJS100[d]	
			Cln	Tet		Plasmid band	Chromosome region
Test strains							
2268	*B. fragilis*	UI[e]	128	32	−	−	+
2300	*B. fragilis*	UI	256	16	−	−	+
2413	*B. fragilis*	UI	256	32	+	+	+
2417	*B. thetaiotaomicron*	UI	256	16	−	−	+
2423	*B. fragilis*	UI	256	32	+	−	+
2424	*B. distasonis*	UI	128	8	−	−	+
2426	*B. fragilis*	UI	256	32	+	−	+
2428	*B. thetaiotaomicron*	UI	256	16	−	−	+
2429	*B. vulgatus*	UI	512	8	−	−	+
2459	*B. fragilis*	UI	256	32	+	+	−
1877	*B. vulgatus*	Netherlands	512	2	+	+	+
1878	*B. thetaiotaomicron*	Netherlands	512	8	−	−	+
1912	*B. thetaiotaomicron*	St. Louis, Mo.	256	4	+	−	+
1913	*B. fragilis*	St. Louis, Mo.	256	4	−	−	+
2480	*B. fragilis*	Boston, Mass.	512	16	−	−	+
2450	*B. fragilis*	Brazil	512	16	−	−	−
Controls							
TM4000	*B. fragilis*	Paris, France	0.012	0.25	−	−	−
JC101	*B. fragilis*	Boston, Mass.	0.012	0.25	−	−	−
TMP10	*B. fragilis*	Los Angeles, Calif.	512	16	+	Source of pBFTM10	

[a] TAL, Tufts Anaerobic Laboratory.
[b] MIC, Minimal inhibitory concentration.
[c] As determined by agarose gel electrophoresis. +, Detected; −, not detected.
[d] For pMJS100, see reference 14.
[e] UI, University of Illinois.

showed homology with pBFTM10 or the *Eco*RI fragment of this plasmid cloned in *E. coli*. Results with seven representative strains and two control strains are presented in Fig. 1. Five of the seven strains (TAL1912, TAL2300, TAL2424, TAL2428, and TAL2480) show homology with sequences present in the chromosomal region. It should be pointed out that for these strains and strains discussed below, the "chromosomal region" may also contain plasmid DNA sequences in addition to chromosomal sequences. One strain, TAL1877 (Fig. 1, lane 5), shows homology with the chromosomal region and, in addition, with a small plasmid; *B. fragilis* TAL2450 (lane 9) failed to show any homology. Seven additional strains (data not shown) had homology within the chromosomal region only (Table 1, strains TAL2268, TAL2417, TAL2423, TAL2426, TAL2429, TAL1878, and TAL1913). Two strains, TAL2413 and TAL1877, showed homology within the chromosomal region and in a plasmid. Strain TAL2459 showed homology with a plasmid only (Table 1).

Transfer experiments. The transfer of Cln[r] and Tet[r] was studied with strains TAL2300, TAL2459, TAL2450, and TAL2480 as donors and strain JC101 as the recipient (Table 2). None of these strains transferred Cln[r] or Tet[r] without antibiotic pretreatment. Strains TAL2300 and TAL2459 transferred both Cln[r] and Tet[r] after tetracycline or clindamycin induction. Strain TAL2450 transferred Cln[r] and Tet[r] after growth in tetracycline (1 μg/ml) before mating; there was no transfer of resistance determinants after growth in clindamycin at 1 μg/ml. Strain TAL2480 failed to transfer either Tet[r] or Cln[r] in three separate experiments after induction with clindamycin or tetracycline.

DISCUSSION

The *B. fragilis* plasmid pBFTM10 and the chimeric plasmid pMJS100 were useful probes for the detection of Cln[r] determinants in the referred Cln[r] *Bacteroides* strains. The plasmid pMJS100 was the preferred probe because it could be easily obtained in a large quantity and labeled to a high specific activity by nick translation. This probe gave positive results with several strains that had inconclusive or negative results with the probe pBFTM10. Questions remaining to be investigated include the relation-

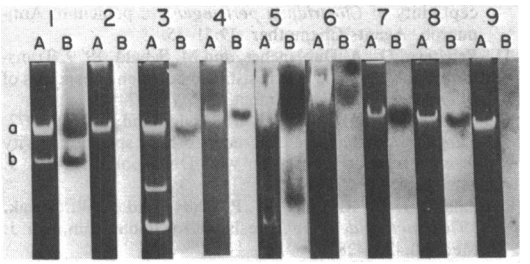

FIG. 1. Agarose gel electrophoresis (A) and Southern blots (B) of seven Cln^r *Bacteroides* strains (lanes 3–9) and two controls (lanes 1 and 2). The figure is a composite of three separate experiments. Hybridization was performed in 0.9 M sodium nitrate–0.2% Ficoll–0.2% polyvinylpyrrolidone–0.5% sodium dodecyl sulfate–0.02% bovine serum albumin V–100 μg of calf thymus DNA per ml at 65°C for 20 h. *Bacteroides* target DNA obtained by the minilysis technique was fractionated by agarose gel electrophoresis and then transferred to nitrocellulose filters. ^{32}P-labeled pBFTM10 or pMJS100 DNA was used as a probe as described in the text. The strains shown are as follows: lane 1, TM4003 (positive control strain which contains pBFTM10 in closed circular form [a] and open circular form [b] [this plasmid has a length of 14.6 kilobases and serves as a size standard]); lane 2, JC101 (negative control); lane 3, TAL1912; lane 4, TAL2300; lane 5, TAL1877; lane 6, TAL2424; lane 7, TAL2428; lane 8, TAL2480; lane 9, TAL2450.

ship of *B. fragilis* Cln^r to other Cln^r anaerobic bacteria, e.g., *Bacteroides* and *Clostridium* spp., and the relationship to the macrolide-lincosamide-streptogramin resistance determinants in *Streptomyces* spp., *Staphylococcus aureus*, and *Bacillus* spp. (4, 5, 23) and to the erythromycin resistance determinant on Tn*917* of *Streptococcus faecalis* (22).

Data from our studies indicate that the Cln^r determinant on pBFTM10 is widely distributed in natural isolates of *Bacteroides* spp. and that the Cln^r determinant may reside on a plasmid or in the chromosome of resistant isolates. These two observations suggest that the Cln^r determinant may be carried on a transposable genetic element. Definitive answers to this question await genetic tools to study transposition in *B. fragilis* or the cloning of the Cln^r determinant in a suitable host for the study of transposition.

The lack of homology between the Cln^r determinant of pBFTM10 and DNA from strain TAL2450 suggests that there may be at least two different clindamycin-erythromycin resistance determinants in *Bacteroides* spp. Further evidence supporting this hypothesis is the failure of clindamycin induction to increase the frequency of transfer in the experiments reported in this paper. We have always observed clindamycin induction to increase the frequency of Cln^r and Tet^r transfer in the Tet^r Cln^r strains previously studied (17, 19). The existence of more than one class of Cln^r determinants in *Bacteroides* spp. is not surprising since multiple determinants leading to resistance to the same antimicrobial agents are often encountered in aerobic bacteria. Multiple determinants have already been demonstrated for erythromycin-clindamycin-streptogramin resistance in *Streptomyces* spp. (5).

The potential for the proliferation of clindamycin resistance in *B. fragilis* exists because of the presence of transfer factors, such as pBFTM10 and pBF4, and the increased use of these drugs in hospitals. pBF4 and pBFTM10 both code for clindamycin and erythromycin resistance. Thus the occurrence of Cln^r *Bacteroides* strains may be enhanced by the use of either clindamycin or erythromycin. The use of erythromycin has increased in the past few years. This drug is used for the treatment of atypical pneumonia caused by *Mycoplasma pneumoniae* or *Legionella pneumophila* and in preoperative prophylactic bowel preparations for colonic surgery. The latter usage occurs in the very patients at high risk to develop infections with *Bacteroides* spp. Currently, there is an outbreak of Cln^r *Bacteroides* spp. on the surgical wards of the University of Illinois Hospital; the epidemiology of this outbreak is being investigated (A. C. England, E. J. Bond, H. Livingston, and K. E. Nelson, personal communication). Based on the above-described data, routine susceptibility testing of *Bacteroides* isolates, particularly for clindamycin susceptibility,

TABLE 2. Transfer frequencies of *B. fragilis* isolates

Donor strain	Recipient strain	Inducing antibiotic	Selected resistance markers	Transfer frequency[a]
TAL2300	JC101	Tet	Tet Rif	1.5×10^{-3}
			Cln Rif	5.77×10^{-4}
		Cln	Tet Rif	0.3×10^{-7}
			Cln Rif	1.2×10^{-7}
TAL2450	JC101	Tet	Tet Rif	8.0×10^{-7}
			Cln Rif	8.0×10^{-7}
		Cln	Tet Rif	$<1 \times 10^{-8}$
			Cln Rif	$<1 \times 10^{-8}$
TAL2459	JC101	Tet	Tet Rif	1.2×10^{-5}
			Cln Rif	1.2×10^{-6}
		Cln	Tet Rif	2.2×10^{-5}
			Cln Rif	1.0×10^{-6}

[a] Expressed as number of transconjugants per number of donor cells input.

is indicated in patients with serious anaerobic infections.

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LITERATURE CITED

1. **Bawdon, R. E., E. Rozmiej, S. Palchaudhuri, and J. Krakowiak.** 1979. Variability in the susceptibility pattern of *Bacteroides fragilis* in four Detroit area hospitals. Antimicrob. Agents Chemother. **16**:664–666.
2. **Casadaban, M. J., J. Chou, and S. N. Cohen.** 1980. In vitro gene fusions that join an enzymatically active β-galactosidase segment to amino-terminal fragments of exogenous proteins: *Escherichia coli* plasmid vectors for the detection and cloning of translational initiation signals. J. Bacteriol. **143**:971–980.
3. **Davis, R. W., D. Botstein, and J. R. Roth.** 1980. A manual for genetic engineering. Advanced bacterial genetics, p. 124. Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.
4. **Docherty, A., G. Grandi, R. Grandi, T. J. Gryczan, A. G. Shivakumar, and D. Dubnau.** 1981. Naturally occurring macrolide-lincosamide-streptogramin B resistance in *Bacillis licheniformis*. J. Bacteriol. **145**:129–137.
5. **Fujisawa, Y., and B. Weisblum.** 1981. A family of r-determinants in *Streptomyces* spp. that specifies inducible resistance to macrolide, lincosamide, and streptogramin type B antibiotics. J. Bacteriol. **146**:621–631.
6. **Gorbach, S. L., and J. G. Bartlett.** 1974. Anaerobic infections. N. Engl. J. Med. **290**:1177–1184, 1237–2354, 1289–1294.
7. **Holdeman, L. V., W. E. C. Moore, M. Rogosa, J. R. Postgate, and M. P. Bryant.** 1974. Gram-negative anaerobic bacteria, p. 384–426. *In* R. E. Buchannan and N. E. Gibbons, (ed.), Bergey's manual of determinative bacteriology, 8th ed. The Williams & Wilkins Co., Baltimore.
8. **Magot, M., F. Fayolle, G. Privitera, and M. Sebald.** 1981. Transposon-like structures in the *Bacteroides fragilis* MLS plasmid pIP410. Mol. Gen. Genet. **181**:559–561.
9. **Malamy, M. H., and F. P. Tally.** 1981. Mechanisms of drug-resistance transfer in *Bacteroides fragilis*. J. Antimicrob. Chemother. **8**(Suppl. D):59–75.
10. **Marrie, T. J., E. V. Haldane, C. A. Swantee, and E. A. Kerr.** 1981. Susceptibility of anaerobic bacteria to nine antimicrobial agents and demonstration of decreased susceptibility of *Clostridium perfringens* to penicillin. Antimicrob. Agents Chemother. **19**:51–55.
11. **Privitera, G., A. Dublanchet, and M. Sebald.** 1979. Transfer of multiple antibiotic resistance between subspecies of *Bacteroides fragilis*. J. Infect. Dis. **139**:97–101.
12. **Rigby, P. W., M. Dieknan, C. Rhodes, and P. Berg.** 1977. Labelling deoxyribonucleic acid to high specific activity *in vitro* by nick translation with DNA polymerase I. J. Mol. Biol. **113**:237–257.
13. **Salaki, J. S., R. Black, F. P. Tally, and J. W. Kislak.** 1976. *Bacteroides fragilis* resistant to clindamycin. Am. J. Med. **60**:426–428.
14. **Shimell, M. J., C. J. Smith, F. P. Tally, F. L. Macrina, and M. H. Malamy.** 1982. Hybridization studies reveals homology between pBF4 and pBFTM10, two clinidamycin-erythromycin resistance transfer plasmids of *Bacteroides fragilis*. J. Bacteriol. **152**:950–953.
15. **Southern, E. M.** 1975. Detection of specific sequences among DNA fragments separated by gel electrophoresis. J. Mol. Biol. **98**:503–517.
16. **Tally, F. P., N. V. Jacobus, and M. Barza.** 1979. *In vitro* susceptibility and serum protein-binding of cefaclor. J. Antimicrob. Chemother. **5**:159–165.
17. **Tally, F. P., M. J. Shimell, G. R. Carson, and M. H. Malamy.** 1981. Chromosomal and plasmid-mediated transfer of clindamycin resistance in *Bacteroides fragilis*, p. 51–60. *In* S. B. Levy, R. C. Clowes, and E. L. Koenig (ed.), Molecular biology, pathogenicity and ecology of bacterial plasmids. Plenum Publishing Corp., New York.
18. **Tally, F. P., D. R. Snydman, S. L. Gorbach, and M. H. Malamy.** 1979. Plasmid mediated, transferable resistance to clindamycin and erythromycin in *Bacteroides fragilis*. J. Infect. Dis. **139**:83–88.
19. **Tally, F. P., D. R. Snydman, M. J. Shimell, and M. H. Malamy.** 1982. Characterization of pBFTM10 a clindamycin-erythromycin resistance transfer factor from *Bacteroides fragilis*. J. Bacteriol. **151**:686–691.
20. **Tally, F. P., A. Sosa, N. V. Jacobus, and M. H. Malamy.** 1981. Clindamycin resistance in *Bacteroides fragilis*. J. Antimicrob. Chemother. **8**(Suppl. D):43–48.
21. **Thadepalli, H., S. L. Gorbach, and P. W. Broido.** 1973. Abdominal trauma, anaerobes, and antibiotics. Surg. Gynecol. Obstet. **137**:270–276.
22. **Tomich, P. K., F. Y. An, and D. B. Clewell.** 1980. Properties of erythromycin-inducible transposon Tn*917* in *Streptococcus faecalis*. J. Bacteriol. **141**:1366–1374.
23. **Weisblum, B., S. B. Holder, and S. M. Halling.** 1979. Deoxyribonucleic acid sequence common to staphylococcal and streptococcal plasmids which specify erythromycin resistance. J. Bacteriol. **138**:990–998.
24. **Welch, R. A., K. R. Jones, and F. L. Macrina.** 1979. Transferable lincosamide-macrolide resistance in *Bacteroides*. Plasmid **2**:261–268.
25. **Welch, R. A., and F. L. Macrina.** 1981. Physical characterization of *Bacteroides fragilis* R plasmid pBF4. J. Bacteriol. **145**:867–872.