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Antimicrobial Resistance among Clinical Isolates of *Streptococcus pneumoniae* in Canada during 2000

Donald E. Low,[1,2]* Joyce de Azavedo,[1,2] Karl Weiss,[3] Tony Mazzulli,[1,2] Magdalena Kuhn,[4] Deirdre Church,[5] Kevin Forward,[6] George Zhanel,[7] Andrew Simor,[8] Canadian Bacterial Surveillance Network,† and A. McGeer[1,2]

Department of Microbiology, Toronto Medical Laboratories and Mount Sinai Hospital,[1] University of Toronto,[2] and Sunnybrooke and Women's College Health Sciences Center,[8] Toronto, Ontario, and Hopital Maisonneuve-Rosemont, University of Montreal, Montreal, Quebec,[3] Southeast Healthcare Corporation-Moncton Site, Moncton, New Brunswick,[4] Calgary Laboratory Services, Calgary, Alberta,[5] Queen Elizabeth II Health Sciences Center, Halifax, Nova Scotia,[6] and Health Sciences Center, Winnipeg, Manitoba,[7] Canada

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A total of 2,245 clinical isolates of *Streptococcus pneumoniae* were collected from 63 microbiology laboratories from across Canada during 2000 and characterized at a central laboratory. Of these isolates, 12.4% were not susceptible to penicillin (penicillin MIC, ≥0.12 μg/ml) and 5.8% were resistant (MIC, ≥2 μg/ml). Resistance rates among non-β-lactam agents were the following: macrolides, 11.1%; clindamycin, 5.7%; chloramphenicol, 2.2%; levofloxacin, 0.9%; gatifloxacin, 0.8%; moxifloxacin, 0.4%; and trimethoprim-sulfamethoxazole, 11.3%. The MICs at which 90% of the isolates were inhibited (MIC_{90}s) of the fluoroquinolones were the following: gemifloxacin, 0.03 μg/ml; BMS-284756, 0.06 μg/ml; moxifloxacin, 0.12 μg/ml; gatifloxacin, 0.25 μg/ml; levofloxacin, 1 μg/ml; and ciprofloxacin, 1 μg/ml. Of 578 isolates from the lower respiratory tract, 21 (3.6%) were inhibited at ciprofloxacin MICs of ≥4 μg/ml. None of the 768 isolates from children were inhibited at ciprofloxacin MICs of ≥4 μg/ml, compared to 3 of 731 (0.6%) from those ages 15 to 64 (all of these >60 years old), and 27 of 707 (3.8%) from those over 65. The MIC_{90}s for ABT-773 and telithromycin were 0.015 μg/ml for macrolide-susceptible isolates and 0.12 and 0.5 μg/ml, respectively, for macrolide-resistant isolates. The MIC of linezolid was ≤2 μg/ml for all isolates. Many of the new antimicrobial agents tested in this study appear to have potential for the treatment of multidrug-resistant strains of pneumococci.

The presence of *Streptococcus pneumoniae* isolates resistant to penicillin and the macrolide antibiotics has raised concerns regarding the use of these antimicrobial agents as empirical agents for the treatment of community-acquired pneumonia (6). Fluoroquinolones with increased activity against *S. pneumoniae*, such as levofloxacin, moxifloxacin, and gatifloxacin, are now being recommended and widely used for the treatment of patients with community-acquired pneumonia who are at risk for infection due to multidrug-resistant strains (6, 19, 22, 33, 41, 44). However, resistance to this class of agents is also emerging (11, 24), and treatment failures have been reported (12). Consequently, in order to track emerging resistance, there is a need to continue surveillance and to monitor resistance trends even with recently developed compounds.

The ketolides and oxazolidinones are two new drug classes that may have a role in the treatment of infections due to *S. pneumoniae*. The ketolides are semisynthetic macrolides made by the replacement of the cladinose at C-3 with a keto group (8). This alteration of the natural erythromycin A molecule renders the drug more stable in acidic environments and reduces the induction of the macrolide-lincosamide-streptogramin B (MLS_B) resistance phenotype (7). Telithromycin (formerly called HMR 3647 or RU 66647) and ABT-773, two new ketolides, have potential for the treatment of pneumococcal infections, including those caused by macrolide-resistant strains, whether possessing the *erm*(B) or the *mef*(A) genotype (1, 14, 18). The oxazolidinones comprise a novel synthetic class of antimicrobial agents with activity against gram-positive aerobes, gram-negative anaerobes, and mycobacteria (51, 59). One member of this class, linezolid, acts by inhibiting the initiation phase of protein translation through direct interaction with the bacterial ribosome and the 70S rRNA initiation complex (49).

We present here the in vitro activity of commonly used antimicrobial agents, as well as the newer fluoroquinolones, the ketolides, and linezolid against *S. pneumoniae* isolates collected in 2000 through an ongoing cross-Canada surveillance study.

MATERIALS AND METHODS

Members of the Canadian Bacterial Surveillance Network, comprising private laboratories and community and university-affiliated hospital laboratories from across Canada, were asked to collect the first 20 consecutive clinical isolates followed by all sterile site isolates of *S. pneumoniae* in 2000. Date of collection, source of specimen, and patient age and gender were recorded on a standardized form. Duplicate isolates from the same patient were excluded. Isolates were transported on chocolate agar slants or swabs to a central laboratory. Upon receipt, the isolates were confirmed as *S. pneumoniae* by standard methodology. Following storage at −70°C, isolates were thawed and subcultured onto blood agar twice before susceptibility testing was performed. In vitro susceptibility testing was performed by broth microdilution and disk diffusion testing according to National Committee for Clinical Laboratory Standards (NCCLS) guidelines

* Corresponding author. Mailing address: Department of Microbiology, Rm. 1487, Mount Sinai Hospital, 600 University Ave., Toronto, Ontario M5G 1X5, Canada. Phone: (416) 586-4435. Fax: (416) 586-8746. E-mail: dlow@mtsinai.on.ca.

† Members are listed in the Appendix.

TABLE 1. In vitro activities of 16 antimicrobial agents against 2,245 isolates of *S. pneumoniae* collected from across Canada during 2000

Antimicrobial agent (no. of isolates tested)[a]	MIC (μg/ml)			% of isolates per category[b]	
	50%	90%	Range	I	R
Penicillin	0.03	0.12	≤0.015–4	6.6	5.8
Amoxicillin	0.03	0.12	≤0.015–8	<1	<1
Ceftriaxone[c]					
Meningitis (44)	≤0.03	0.5	≤0.03–2	6.8	2.3
Nonmeningitis (2,201)	≤0.03	0.5	≤0.03–8	1.9	0.1
Ciprofloxacin	1	1	≤0.12–64	3	1.4
Levofloxacin	1	1	≤0.12–32	<1	0.9
Gatifloxacin	0.25	0.25	≤0.03–16	<1	<1
Moxifloxacin	0.12	0.12	≤0.03–8	<1	<1
BMS-284756	0.03	0.06	≤0.015–2	NA[d]	NA
Gemifloxacin	0.015	0.03	≤0.007–2	NA	NA
Doxycycline (2,143)	≤1	1	≤1–16	NA	NA
Chloramphenicol	≤4	4	≤4–≥16	NA	2.2
Erythromycin	≤0.12	2	≤0.12–≥128	<1	11.1
Clindamycin	≤0.25	0.25	≤0.25–≥128	<1	5.5
Telithromycin[e]					
MS	≤0.015	0.015	≤0.015–0.5	NA	NA
MR	0.06	0.5	≤0.015–2	NA	NA
ABT-773[e]					
MS	≤0.015	0.015	≤0.015–0.03	NA	NA
MR	0.03	0.12	≤0.015–1	NA	NA
Linezolid (2143)	1	1	0.25–2	NA	NA
TMP/SMX	≤0.25	4	≤0.25–≥128	10.5	11.3

[a] Number of isolates tested was 2,245 unless otherwise indicated.
[b] I, intermediate; R, resistant.
[c] The NCCLS susceptibility interpretive criteria for ceftriaxone for meningeal and nonmeningeal isolates of *S. pneumoniae* (40).
[d] NA, not applicable.
[e] MS, macrolide susceptible; MR, macrolide resistant.

(38, 39). Susceptibility interpretive criteria used were those published in the NCCLS M100-S12 document (40). The nonsusceptible category was defined as those isolates with MICs in the intermediate and resistant category. The NCCLS M100-S12 guidelines have included new interpretive criteria for nonmeningeal isolates of *S. pneumoniae* for cefotaxime and ceftriaxone: drug MICs of ≤1, 2, and ≥4 μg/ml for susceptible, intermediate, and resistant, respectively. The current absence of data on strains resistant to linezolid precludes defining any categories other than susceptible (MIC, ≤2 μg/ml). For the purpose of this study, ciprofloxacin MICs of ≥4 μg/ml were used to define the resistance category. An MIC of ≥4 μg/ml was chosen because of the association of this degree of resistance with mutations in the quinolone resistance-determining regions of genes encoding DNA topoisomerase IV and DNA gyrase A (28, 46, 55). The antimicrobial agents were supplied by their respective manufacturers or purchased from Sigma (Sigma, St. Louis, Mo.). Erythromycin-resistant isolates were further classified as having either the M or MLS_B phenotype. Isolates that were erythromycin resistant and clindamycin susceptible were classified as having the M phenotype, and those that were both erythromycin and clindamycin resistant were classified as having the MLS_B phenotype (29, 32, 52). All erythromycin-resistant isolates (by broth microdilution) were tested using a double-diffusion disk test, as described elsewhere (43, 48), with erythromycin (15 μg) and clindamycin (2 μg) disks placed 20 mm apart on 5% defibrinated horse blood agar and incubated overnight at 35°C in a 5% carbon dioxide atmosphere. After incubation, the presence or absence of blunting in the zone of inhibition of the clindamycin disk was recorded. If the clindamycin inhibition zone was blunted toward the erythromycin disk, the strain was interpreted as clindamycin inducible.

RESULTS

A total of 2,245 isolates of pneumococci were collected from 63 clinical microbiology laboratories across Canada. Of these, 901 (40%) were isolated from blood cultures, 578 (24%) from lower respiratory tract specimens, 439 (20%) from conjunctival swabs, 217 (10%) from ear swabs, 44 (2%) from cerebrospinal fluid, and 66 (2%) from other sites. Of the 2,203 isolates for which ages of patients were available, 768 (35%) were from patients <15 years old, 731 (33%) were from patients between 15 and 64 years old, and 707 (32%) were from patients ≥65 years old. The results of in vitro susceptibility testing are found in Tables 1 and 2. In this study, we found that 12.4% of *S. pneumoniae* isolates were penicillin nonsusceptible, 6.6% fell in the intermediate category (penicillin MIC, 0.12 to 1 μg/ml), and 5.8% were penicillin resistant (MIC, ≥2 μg/ml) (Table 1). Forty-four *S. pneumoniae* isolates were taken from cerebrospinal fluid. Using meningeal interpretive criteria, three (6.8%) were intermediate and one (2.3%) was resistant to ceftriaxone. There were 2,201 nonmeningeal isolates. Using the nonmeningeal interpretive criteria, 42 (1.9%) were intermediate and 2 (0.1%) were resistant to ceftriaxone.

Recently, pneumococci with high-level resistance to amoxicillin (amoxicillin MICs, ≥4 μg/ml) and/or extended-spectrum cephalosporins (cefotaxime MICs, ≥4 μg/ml) have been identified in France (15). The emergence of such strains is of considerable concern because it further limits the available options for the therapy of severe pneumococcal infections. We identified only 10 isolates (0.4%) for which amoxicillin MICs were 4 μg/ml and one for which the MIC was 8 μg/ml. Only two isolates, for each of which the amoxicillin MIC was 2 μg/ml, had ceftriaxone MICs of ≥4 μg/ml (Table 2).

A total of 249 (11.1%) isolates were macrolide resistant, of which 128 (51%) were clindamycin resistant. Therefore, 51% of macrolide-resistant strains had the MLS_B phenotype and 49% had the M phenotype. Two (0.8%) isolates had an induc-

TABLE 2. In vitro activities of 16 antimicrobial agents against 2,245 isolates of *S. pneumoniae* collected from across Canada during 2000

Antimicrobial agent	No. of isolates inhibited by MIC (μg/ml) of[a]:														
	0.007	0.015	0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	64	128
Penicillin		586[b]	1,233	147	75	18	18	38	92	38					
Amoxicillin		84[b]	1,153	734	73	37	46	74	33	10	1				
Ceftriaxone			1,824[b]	60	41	40	126	109	43	1	1				
Ciprofloxacin					2[b]	63	892	1,188	68	7	7	6	9	3[c]	
Levofloxacin					1[b]	19	832	1,355	16	2	5	12	3		
Gatifloxacin			1[b]	19	548	1,612	40	4	3	14	3	1			
Moxifloxacin			25[b]	449	1,675	74	1	0	12	8	1				
BMS-284756	44[b]	294	1,533	344	9	4	10	6	1						
Gemifloxacin	588[b]	1,214	401	20	3	11	6	1	1						
Doxycycline[d]								1,960[b]	10	91	72	10			
Chloram[e]										2,198[b]	2	45[c]			
Erythromycin					1,984[b]	9	3	10	28	42	28	18	12	24	87[c]
Clindamycin						2,118[b]	3	0	4	4	2	5	6	13	90[c]
Telithromycin		≥2,016[b]	59	49	37	23	40	19	2						
ABT-773		2,102[b]	49	40	39	12	2	1							
Linezolid[d]						29	966	1,054	94						
TMP/SMX						1,501[b]	254	149	87	85	144	24	0	0	1[c]

[a] Underlined number denotes intermediate category where applicable.
[b] MICs for these isolates were less than or equal to the value given.
[c] MICs for these isolates were greater than or equal to the value given.
[d] Only 2,143 isolates were tested.
[e] Chloramphenicol.

ible phenotype. Both of these isolates were resistant by zone diameter. For only four isolates did the broth microdilution susceptibility testing results for clindamycin not correlate with the disk diffusion susceptibility testing results. Three isolates were susceptible, and one was intermediate, by broth microdilution testing but resistant by disk diffusion. Two of the susceptible isolates were inducible by disk diffusion testing. Sixty-six percent of macrolide-resistant isolates from sterile sites had the M phenotype, versus 40% from nonsterile sites ($P < 0.001$). Penicillin and macrolide resistance were evenly distributed in the three age groups.

More than 99% of isolates tested had telithromycin and ABT-773 MICs of ≤1 μg/ml (Table 2). The MIC_{50}s and MIC_{90}s of telithromycin and ABT-773 for macrolide-susceptible isolates were both ≤0.015 μg/ml (Table 1). The MIC_{50}s and MIC_{90}s of telithromycin and ABT-773 for macrolide-resistant isolates were 0.06 and 0.5 μg/ml for telithromycin and 0.03 and 0.12 μg/ml for ABT-773, respectively. MICs of telithromycin and ABT-773 were higher for isolates with the M phenotype than for those with the MLS_B phenotype. Of macrolide-resistant isolates with the M phenotype, for 43 of 121 (36%) the telithromycin MIC was ≥0.5 μg/ml and for 38 of 121 (31%) the ABT-773 MIC was ≥0.125 μg/ml. In comparison, among isolates with the MLS_B phenotype, only 16 of 128 (13%) had a telithromycin MIC of ≥0.5 μg/ml and 13 of 128 (10%) had an ABT-773 MIC of ≥0.125 μg/ml ($P < 0.001$ for each M-versus-MLS_B phenotype comparison). There were 22 isolates for which telithromycin MICs were 1 (20) or 2 (2) μg/ml. Sixteen (73%) had an M phenotype. The MIC_{50} and MIC_{90} of linezolid were both 1 μg/ml. Increased linezolid MICs were associated with resistance to penicillin and trimethoprim-sulfamethoxazole (TMP/SMX) but not macrolides or tetracycline. Twenty-three of ninety-four (24%) isolates with linezolid MICs of 2 μg/ml were resistant to penicillin, compared to 105 of 2,049 (5%) susceptible isolates ($P < 0.001$). Thirty of ninety-four (32%) isolates with linezolid MICs of 2 μg/ml were resistant to cotrimoxazole, compared to 220 of 2,049 (11%) susceptible isolates ($P < 0.001$).

Thirty-two (1.4%) isolates were resistant to ciprofloxacin (Table 3). None of 768 isolates from children were ciprofloxacin resistant, compared to 3 of 731 (0.6%) from those ages 15 to 64 (all of these >60 years old) and 27 of 707 (3.8%) from

TABLE 3. The distribution of the MICs of selected fluoroquinolones for 32 *S. pneumoniae* isolates for which ciprofloxacin MICs were ≥4 μg/ml

Antimicrobial agent	No. of isolates inhibited by MIC (μg/ml) of[a]:												
	0.015	0.03	0.06	0.12	0.25	0.5	1	2	4	8	16	32	≥64
Ciprofloxacin									7	7	6	9	3
Levofloxacin							5	5	2	5	12	3	
Gatifloxacin					1	8	2	3	14	3	1		
Moxifloxacin				2	9	0	0	12	8	1			
BMS-284756		1	6	4	4	10	6	1					
Gemifloxacin		2	8	3	11	6	1	1					

[a] Underlined number denotes intermediate category where applicable.

those over 65. The age of the patient was not known for one ciprofloxacin-resistant isolate from sputum. Twenty-one of 578 (3.6%) isolates from the lower respiratory tract were ciprofloxacin resistant, compared to 7 of 901 (0.8%) from blood and 4 of 766 (0.5%) from other sites ($P < 0.001$). One isolate was in the penicillin-intermediate category, and four (12%) were in the resistant category. MIC_{90}s for the fluoroquinolones tested against the 32 ciprofloxacin-resistant isolates were the following: ciprofloxacin, 32 μg/ml; levofloxacin, 16 μg/ml; gatifloxacin, 8 μg/ml; moxifloxacin, 4 μg/ml; BMS-284756, 1 μg/ml; and gemifloxacin, 0.5 μg/ml.

DISCUSSION

The results of surveillance studies performed by the Canadian Bacterial Surveillance Network in 1994–95 and 1997–98 have been reported previously (11, 50). The prevalences of pneumococcal isolates that were not susceptible to penicillin, 11.7% in 1994–95 and 13.9% in 1997–98, have remained stable taking into account the rate of 12.7% found in this study ($P = 0.07$). Similarly, the rates of resistance to TMP/SMX of 11.6% in 1997/98 and 11.3% in 2000 have not changed significantly ($P = 0.07$). However, the rates of resistance to the macrolides have shown a temporal increase, ranging from 2.8% in 1994–95 to 6.7% in 1997–98 to 11.6% in 2000 ($P < 0.001$, Chi-square test for trend). By comparison, the prevalence of resistance to not only the macrolides but also the penicillins and TMP/SMX increased in the United States during this time span and is now reported to be in excess of 20% (14, 57). Doern et al. (14) found the overall national rate of nonsusceptible pneumococci to be 34.2%, of which 21.5% were resistant. Macrolide and TMP/SMX resistance increased from 10.3 and 24.8% in 1994–1995 to 26.2 and 35.9% in 1999 to 2000. Although such differences between two countries cannot be easily explained, they are not surprising. Whitney et al. (57) found similar variances in the rates of resistance in different regions of the United States. The proportion of penicillin-resistant isolates was highest in the southeastern region (~34%) and lowest in California (15%) and New York (15%).

In January of 2002, the NCCLS published new susceptibility interpretive criteria for cefotaxime and ceftriaxone for nonmeningeal isolates of *S. pneumoniae*. Previously, susceptibility interpretive breakpoints were ≤0.5, 1, and ≥2 μg/ml for susceptible, intermediate, and resistant, respectively. However, these breakpoints were derived largely from considerations in the treatment of meningitis. The Subcommittee on Antimicrobial Susceptibility Testing (SAST) of NCCLS reviewed surveillance susceptibility data, pharmacokinetic-pharmacodynamic data in animal models of infection, clinical data, and a simulated trial evaluating the probability of attaining target levels in serum relative to various MICs (Mary Jane Ferraro, Chairholder, SAST). As a result of this review, the SAST agreed to publish new interpretive criteria for nonmeningeal isolates of *S. pneumoniae.* The changes are largely designed to clarify the efficacy of these drugs for nonmeningeal infections that are due to *S. pneumoniae* strains with MICs of ≤1 μg/ml. Thus, new interpretive criteria for susceptibility to cefotaxime and ceftriaxone for *S. pneumoniae* in nonmeningitis cases are MICs of ≤1, 2, and ≥4 μg/ml for susceptible, intermediate, and resistant, respectively (40).

Many of the efforts to control resistance, some of which have been successful, have been aimed at reducing the total consumption of antibiotics. However, establishing a precise quantitative relationship between the frequency of resistance to a defined antibiotic and the volume of drug use has proved to be difficult because of the paucity of longitudinal studies that record resistance and drug use patterns (3, 30). In Canada, there has been an overall reduction in the use of antibiotics in the outpatient setting with the greatest reduction seen in the use of oral β-lactams and TMP/SMX. There has been an overall reduction in the number of antibiotic prescriptions between 1995 and 2000 from 96.5 to 77.7 per 100 patients per year (IMS HEALTH, Canada, unpublished data). During the same time period, there was a reduction in the number of erythromycin prescriptions from 11.1 to 3.8 per 100 patients per year and an increase in the number of clarithromycin and azithromycin prescriptions from 4.1 to 12.1 per 100 patients per year. Hyde and colleagues (26) compared trends in macrolide use and resistance in pneumococci as part of an ongoing surveillance program carried out in the United States. From 1993 to 1999, macrolide use increased 13%. Macrolide resistance increased from 10.6% in 1995 to 20.4% in 1999 and could be accounted for by an increase in M phenotypes, while the proportion with the MLS_B phenotype remained stable. Pihlajamaki and colleagues (45), in examining the connection between antimicrobial resistance of pneumococci and regional use of antimicrobial agents in Finland, found that resistance to macrolides and TMP/SMX correlated significantly with the use of these drugs. However, no correlation was found between penicillin resistance and the use of any antimicrobial agent. Arason et al. (2) studied the correlation of antimicrobial consumption with the carriage rate of penicillin-resistant and multiresistant pneumococci in children in Iceland. They found by multivariate analysis that age (<2 years), area (highest antimicrobial consumption), and individual use of antimicrobial agents significantly influenced the odds of carrying penicillin-resistant pneumococci. By univariate analysis, recent antimicrobial use (2 to 7 weeks) and use of TMP/SMX were also significantly associated with carriage of penicillin-resistant pneumococci. Similar findings were made in Sweden, where studies of individual patients showed that TMP/SMX use was the most important predictor of penicillin resistance (35).

In this study, we used erythromycin and clindamycin susceptibility phenotypes to imply resistant mechanisms. Others have shown that phenotypic expression can be used to distinguish between target site modification (*erm* gene mediated; MLS_B phenotype) and efflux resistance mechanisms (*mef* gene mediated; M phenotype) in the majority of *S. pneumoniae* macrolide-resistant isolates (29, 32, 52). Other resistance mechanisms exist but are uncommon (25, 29, 36, 37, 47). Although susceptibility testing may reliably distinguish the M from the MLS_B phenotype in pneumococci, it cannot distinguish those strains with *erm*(B) from those harboring both *erm*(B) and *mef*(A) resistance determinants (27, 34, 36). However, previous studies in Canada have found that the prevalence of macrolide-resistant pneumococci with both *erm*(B) and *mef*(A) resistance determinants is <3% (25, 29, 50). Fasola et al. (17) reported that 43% of erythromycin-resistant pneumococci appeared susceptible to clindamycin at 24 h by broth microdilution when incubated in ambient air but were resistant when tested by disk

diffusion according to NCCLS guidelines. However, when we tested isolates that were found by broth microdilution to be erythromycin resistant and clindamycin susceptible, by disk diffusion, only 3 of 125 (2.4%) isolates were clindamycin resistant. These results support observations made by other large surveillance studies regarding the prevalence of M and MLS_B phenotypes, where only broth microdilution testing was done (14, 26).

Ketolides retain activity against strains of *S. pneumoniae* that have become resistant to macrolides as a result of efflux or target site modification (9, 37). The improved activity of the ketolides is thought to be due to the different modes of action of the ketolides compared with the macrolides, most notably in terms of the strength and nature of their ribosomal binding (16). Macrolides and ketolides bind to two sites within the bacterial ribosome, domains II and V of 23S rRNA, with the interaction at domain II being relatively weak with the macrolides compared to the ketolides. In contrast to the macrolides, the ketolides retain in part their ability to bind to macrolide-resistant pneumococci with the MLS_B phenotype owing to their strong interaction with domain II (16). More than 98% of pneumococci, including macrolide-resistant strains, were inhibited by ≤0.5 μg of telithromycin/ml (31, 42). However, there have been other reports of an increase in the MICs of the ketolides for macrolide-resistant strains (5, 9, 13, 21, 25, 37). We also found that there was a reduction in ketolide activity against macrolide-resistant strains and that the reduction was significantly greater for isolates with the M phenotype than for those with the MLS_B phenotype. Although the MICs for the great majority of such isolates still fall into the susceptible category, there could be selective pressure for development of additional resistance mechanisms with the introduction of these agents into general use, which may be additive and result in elevated MICs that fall in the resistant category (53). For example, in addition to the known mechanisms of macrolide resistance, amino acid substitutions in the large-subunit ribosomal proteins L4 and L22 adjacent to the binding sites of macrolides and ketolides in domains II and IV, respectively, have also been shown to contribute to resistance (4, 54, 58) by altering the conformation of the drug binding site (16). Two previously identified ketolide-resistant isolates from Toronto have been characterized (53; A. G. Tait-Kamradt, R. R. Reinert, A. Al-Lahham, D. E. Low, and J. A. Sutcliffe, Abstr. 41st Intersci. Conf. Antimicrob. Agents Chemother., abstr. 1813, 2001)). For one isolate, the telithromycin MIC was 3.12 μg/ml, but the isolate had no known mechanisms of macrolide resistance other than an L4 insertion (53). For the other strain, isolated from the conjunctiva of a 1-year-old boy in 1996, telithromycin and ABT-773 MICs were 256 and 64 μg/ml, respectively, and the strain was found both to harbor the *erm*B determinant and to contain substitutions in L4 identical to those of several clones that have already been described (53; Tait-Kamradt et al., 41st ICAAC)).

Linezolid has demonstrated MIC_{90}s of ≤2 μg/ml for pneumococci tested (14, 20, 23). Reservoirs of linezolid resistance are less likely, since no analogue has been used previously. Although mutational resistance is extremely difficult to select for in vitro, resistance to linezolid has been reported to have emerged in one isolate of methicillin-resistant *Staphylococcus aureus* (56) and two isolates of *Enterococcus faecium* (G. E. Zurenko, W. M. Todd, B. Hafkin, et al., Abstr. 39th Intersci. Conf. Antimicrob. Agents Chemother., abstr. 848, 1999) recovered from patients during prolonged therapy with linezolid. The mechanism entailed mutations within rRNA genes. Bacteria carry multiple copies of these genes, and changes to single copies may not give rise to phenotypic changes; resistance may be evident only when multiple copies are affected. Thus, selection for resistance might require several steps. Although unexplained, it was interesting that a reduction in linezolid activity was significantly associated with a reduction in both penicillin and TMP/SMX activity.

The prevalence of pneumococci with reduced susceptibility to ciprofloxacin in Canada remained similar to what was reported from surveillance studies carried out in 1997 and 1998: 1.7% overall and 2.9% in adults (11). The 1.4% ciprofloxacin resistance rates are identical to those reported by Doern et al. (14) in their surveillance study carried out over the same time period. Although ciprofloxacin resistance rates were not reported by the Centers for Disease Control's Active Bacterial Core Surveillance (ABCs) during 1995–1999 (10), they did find levofloxacin resistance rates of 0.2%, similar to those of 0.7% reported here. Both reports suggest that rates of fluoroquinolone resistance in the United States are now the same as those reported in Canada. Both this study and the ABCs study found that older patients with respiratory tract infections are at greater risk for colonization and infection with a fluoroquinolone-resistant strain. It may be prudent to now perform routine fluoroquinolone susceptibility testing on pneumococci isolated from the lower respiratory tract in this group of patients. Levofloxacin, gatifloxacin, and moxifloxacin demonstrated excellent in vitro activity. However, gemifloxacin and BMS-284756 were from 4- to 32-fold more active than these agents against ciprofloxacin-susceptible and -resistant pneumococci.

In conclusion, although older agents used to treat pneumococcal infections show reduced activity, the new antimicrobial agents tested in this study appear to have potential for the treatment of these multidrug-resistant strains.

APPENDIX

Members of the Canadian Bacterial Surveillance Network and their participating laboratories were the following: K. Green and S. Porter-Pong, Toronto Medical Laboratories and Mount Sinai Hospital, Toronto, Ontario; H. R. Devlin, St. Michael's Hospital, Toronto, Ontario; R. G. Lewis, Cape Breton Regional Health Care Complex, Sydney, Nova Scotia; P. C. Kibsey, Victoria General Hospital, Victoria, British Columbia; J. Blondeau, Royal University Hospital, Saskatoon, Saskatchewan; W. Geddie, Credit Valley Hospital, Toronto, Ontario; G. K. Harding, St. Boniface General Hospital, Winnipeg, Manitoba; D. Hoban, Health Sciences Centre, Winnipeg, Manitoba; L. Thibault, Hopital Georges L. Dumont, Moncton, New Brunswick; F. Smaill, Hamilton Health Sciences Corporation, Chedoke-McMaster, Hamilton, Ontario; M. Gourdeau and G. Murray, Hopital de l'Enfant-Jesus, Laval University, Quebec City, Quebec; S. Richardson, Hospital for Sick Children, Toronto, Ontario; G. J. Hardy, Saint John Regional Hospital, Saint John, New Brunswick; P. R. Laberge, Centre Hospitalier Regional de Sept-Iles, Sept-Iles, Quebec; L. P. Abbott, Queen Elizabeth Hospital, Charlottetown, Prince Edward Island; M. Yorke, Westman Regional Laboratory, Brandon, Manitoba; N. Clerk, William Osler Health Center-Etobicoke Campus, Toronto, Ontario; J. Downey, Toronto East General and Orthopedic Hospital Inc., Toronto, Ontario; M. Bergeron, CHUQ-Ctr Hopital, Université Laval, Sainte-Foy, Quebec; and G. S. Randhawa, Kelowna General Hospital, Kelowna, British Columbia; J. Hutchinson, Health Care Corporation,

St. John's, Newfoundland; S. Krajden, St. Joseph's Health Care Center, Toronto, Ontario; R. Price, Royal Victoria Hospital, Barrie, Ontario; J. F. Paradis, Hopital de Chicoutimi, Chicoutimi, Quebec; L. Bocci, Regional Hospital Centre, Bathurst, New Brunswick; M. Savard, David Thompson Regional Laboratories, Red Deer, Alberta; B. F. Rudrick, Grey Bruce Regional Hospital, Owen Sound, Ontario; Ostrowska, Trillium Health Centre, Mississauga, Ontario; P. Pieroni, Provincial Laboratory, Regina, Saskatchewan; B. Mederski, North York General Hospital, Toronto, Ontario; C. Tremblay, Hotel Dieu de Quebec, Quebec City, Quebec; P. Leighton, Everett Chalmers Hospital, Fredericton, Nova Scotia.

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