Clonal Relatedness of Erythromycin-Resistant *Streptococcus pyogenes* Isolates in Germany

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In a nationwide study in Germany, a total of 381 *Streptococcus pyogenes* were collected. Erythromycin A-resistant strains were characterized for the underlying resistance genotype, showing 55.6% had the efflux type *mef*(A), 31.5% had *erm*(A), and 13.0% had *erm*(B). A total of 23 different multilocus sequence types were observed.

Streptococcus pyogenes is a major cause of human disease, producing both mild (e.g., pharyngitis) and severe (e.g., toxic shock-like syndrome, necrotizing fasciitis) infections (6). Macrolides are treatment alternatives to penicillin, valuable especially in patients allergic to β -lactams. Different phenotypes of macrolide resistance have been recognized. Strains of the M phenotype are resistant to 14- and 15-member macrolides, due to the presence of *mef*(A) genes, conferring efflux of the antibiotic out of the bacterial cell. These strains remain susceptible to 16-member ring macrolides and clindamycin (16).

Target site modification is mostly based on dimethylation of an adenine residue (A2058) of the 23S rRNA domain V through the action of a family of enzymes encoded by *erm* class genes. Two classes of methylase genes, *erm*(B) and *erm*(A), subclass *erm*(TR), have been described (15). Some of the constitutively resistant *erm*(B)-positive *S. pyogenes* strains were found to be telithromycin resistant (13), but telithromycin retains activity against strains possessing the other macrolide resistance genotypes, such as *erm*(A) and *mef*(A).

In the present study, consecutive clinical isolates ($n = 381$) were collected between November 1999 and March 2000 from outpatients with *S. pyogenes* infections. *S. pyogenes* isolates were identified by hemolysis on sheep blood agar; Lancefield grouping, using a commercially available agglutination technique (Slidex, Streptokit; BioMérieux, Marcy-L'Etoile, France); and a positive pyrrolidonyl-arylamidase test. MIC testing was performed by the broth microdilution method as recommended by the National Committee for Clinical Laboratory Standards (NCCLS) (10). *Streptococcus pneumoniae* ATCC 49619 was used as a control strain. Current NCCLS interpretive criteria were used to define antimicrobial resistance (11). In the case of telithromycin, breakpoints proposed by Fuchs et al. (8) of ≤ 1 and ≥ 4 mg/ml were used for susceptible and

resistant category, respectively. NCCLS breakpoints for roxithromycin were not available. The macrolide resistance phenotype was determined on the basis of the pattern of susceptibility to macrolide-lincosamide-streptogramin B (MLS) antibiotics and confirmed on the basis of the double-disk (erythromycin A plus clindamycin) agar diffusion test (9).

Macrolide-resistant *S. pyogenes* strains were tested by PCR for the presence of *erm*(A), *erm*(B), or *mef*(A) (13, 14).

The two telithromycin-resistant strains were screened for further resistance determinants, including *ere*(A), *ere*(B), *msr*(A), *mph*(A), *mph*(B), and *mph*(C), as described previously (16, 17). The primers and PCR conditions used to amplify 23S rRNA were identical to those used to amplify the *S. pneumoniae* gene (17). *emm* typing was performed as described by Podbielski et al. (12). Multilocus sequence typing was performed as described by Enright et al. for all macrolide-resistant strains (7).

S. pyogenes isolates ($n = 381$) were predominantly isolated from the throat ($n = 256$ [67.2%]) and skin ($n = 83$ [21.8%]). Thirteen strains were isolated from the ear, and 29 were isolated from other sources. Data on antibiotic resistance of all strains and macrolide-resistant strains are presented in Table 1. Resistance to erythromycin A was detected in 54 strains (14.2%). One strain (MSR 141) was levofloxacin-resistant (MIC, ≥ 32 μ g/ml), and two strains were resistant to telithromycin. Additional data on macrolide-resistant strains are presented in Table 2. *S. pyogenes* strains belonged to the following resistance genotypes: *mef*(A) ($n = 30$, 55.6%), *erm*(A) ($n = 17$, 31.5%), and *erm*(B) ($n = 7$, 13.0%). All *mef*(A)-positive strains showed the M phenotype as well as a slightly elevated telithromycin MIC at which 90% of the isolates tested are inhibited (MIC₉₀). All *erm*(A)-positive strains showed inducible resistance to clindamycin. *erm*(B)-positive strains were inducibly clindamycin resistant or showed the cMLS_B phenotype. Two of the *erm*(B)-positive strains were telithromycin resistant. The two telithromycin-resistant strains (MSR 63, telithromycin MIC, 32 μ g/ml; MSR 610, telithromycin MIC, 4 μ g/ml; both cMLS_B phenotypes) did not contain additional macrolide resistance mechanisms, and no mutations were present in L4 or L22 ribosomal proteins or in the 23S rRNA sequences. In

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TABLE 1. Antimicrobial susceptibility of *S. pyogenes* isolates with different macrolide resistance genotypes in Germany from 1999 to 2000

<i>S. pyogenes</i> type (<i>n</i>)	Antibiotic	MIC ($\mu\text{g/ml}$) ^a			No. (%) of strains ^b		
		Range	50%	90%	Susceptible	Intermediate	Resistant
All strains (381)	Penicillin G	≤ 0.016 –0.06	≤ 0.016	≤ 0.016	381 (100)	0 (0)	0 (0)
	Erythromycin A	≤ 0.03 – ≥ 32	≤ 0.03	4	326 (85.6)	1 (0.3)	54 (14.2)
	Clarithromycin	≤ 0.03 – ≥ 32	≤ 0.03	2	327 (85.8)	5 (1.3)	49 (12.9)
	Roxithromycin ^c	≤ 0.03 – ≥ 32	0.125	8	ND ^d	ND	ND
	Azithromycin	≤ 0.03 – ≥ 32	0.06	8	324 (85.0)	2 (0.5)	55 (14.4)
	Clindamycin	≤ 0.03 – ≥ 32	≤ 0.03	0.25	377 (98.9)	0 (0)	4 (1.1)
	Levofloxacin	0.125– ≥ 32	0.5	0.5	380 (99.7)	0 (0)	1 (0.3)
	Telithromycin ^b	≤ 0.03 – ≥ 32	≤ 0.03	≤ 0.03	379 (99.5)	0 (0)	2 (0.5)
Erythromycin A resistant (54)	Penicillin G	≤ 0.016 –0.06	≤ 0.016	≤ 0.016	54 (100)	0 (0)	0 (0)
	Erythromycin A	1– ≥ 32	8	≥ 32	0 (0)	0 (0)	54 (100)
	Clarithromycin	0.5– ≥ 32	4	≥ 32	0 (0)	5 (9.3)	49 (90.7)
	Roxithromycin ^c	1– ≥ 32	16	≥ 32	ND	ND	ND
	Azithromycin	1– ≥ 32	8	≥ 32	0 (0)	1 (1.9)	53 (98.1)
	Clindamycin	≤ 0.03 – ≥ 32	0.06	0.25	50 (92.6)	0 (0)	4 (7.4)
	Levofloxacin	0.125–1	0.5	0.5	54 (100)	0 (0)	0 (0)
	Telithromycin ^c	≤ 0.03 – ≥ 32	0.25	0.5	52 (96.3)	0 (0)	2 (3.7)
<i>mef(A)</i> positive (30)	Penicillin G	≤ 0.016	≤ 0.016	≤ 0.016	30 (100)	0 (0)	0 (0)
	Erythromycin A	1–16	8	16	0 (0)	0 (0)	30 (100)
	Clarithromycin	0.5–16	4	8	0 (0)	1 (3.3)	29 (96.7)
	Roxithromycin ^c	1–32	16	32	ND	ND	ND
	Azithromycin	1–32	8	16	0 (0)	1 (3.3)	29 (96.7)
	Clindamycin	≤ 0.03 –0.25	≤ 0.03	0.25	30 (100)	0 (0)	0 (0)
	Levofloxacin	0.125–1	0.5	0.5	30 (100)	0 (0)	0 (0)
	Telithromycin ^c	≤ 0.03 –1	0.25	0.5	30 (100)	0 (0)	0 (0)
<i>erm(A)</i> positive (17)	Penicillin G	≤ 0.016 –0.03	≤ 0.016	≤ 0.016	17 (100)	0 (0)	0 (0)
	Erythromycin A	2– ≥ 32	4	8	0 (0)	0 (0)	17 (100)
	Clarithromycin	0.5– ≥ 32	1	8	0 (0)	4 (23.5)	13 (76.5)
	Roxithromycin ^c	2– ≥ 32	8	32	0 (0)	0 (0)	17 (100)
	Azithromycin	2– ≥ 32	16	32	0 (0)	0 (0)	17 (100)
	Clindamycin ^c	≤ 0.03 –0.25	0.06	0.25	17 (100)	0 (0)	0 (0)
	Levofloxacin	0.25–1	0.25	0.5	17 (100)	0 (0)	0 (0)
	Telithromycin ^c	≤ 0.03	≤ 0.03	≤ 0.03	17 (100)	0 (0)	0 (0)
<i>erm(B)</i> positive (7)	Penicillin G	≤ 0.016	≤ 0.016	≤ 0.016	7 (100)	0 (0)	0 (0)
	Erythromycin A	8– ≥ 32	≥ 32	≥ 32	0 (0)	0 (0)	7 (100)
	Clarithromycin	1– ≥ 32	≥ 32	≥ 32	0 (0)	1 (14.3)	6 (85.7)
	Roxithromycin ^c	4– ≥ 32	≥ 32	≥ 32	0 (0)	0 (0)	7 (100)
	Azithromycin	8– ≥ 32	≥ 32	≥ 32	0 (0)	0 (0)	7 (100)
	Clindamycin	0.25– ≥ 32	≥ 32	≥ 32	3 (42.9)	0 (0)	4 (57.1)
	Levofloxacin	0.25–0.5	0.25	0.5	7 (100)	0 (0)	0 (0)
	Telithromycin ^c	≤ 0.03 – ≥ 32	0.5	≥ 32	5 (71.4)	0 (0)	2 (28.6)

^a 50% and 90%, MIC₅₀ and MIC₉₀, respectively.^b Breakpoints: penicillin G, intermediate, 0.1 to 1 $\mu\text{g/ml}$, resistant, ≥ 2 $\mu\text{g/ml}$; erythromycin A, intermediate, 0.5 $\mu\text{g/ml}$, resistant, ≥ 1 $\mu\text{g/ml}$; clarithromycin, intermediate, 0.5 $\mu\text{g/ml}$, resistant, ≥ 1 $\mu\text{g/ml}$; azithromycin, intermediate, 1 $\mu\text{g/ml}$, resistant, ≥ 2 $\mu\text{g/ml}$; clindamycin, intermediate, 0.5 $\mu\text{g/ml}$, resistant, ≥ 1 $\mu\text{g/ml}$; and levofloxacin, intermediate, 4 $\mu\text{g/ml}$, resistant, ≥ 8 $\mu\text{g/ml}$ (11).^c Roxithromycin breakpoints are not NCCLS approved. For telithromycin breakpoints of ≤ 1 $\mu\text{g/ml}$, ≥ 4 $\mu\text{g/ml}$ was used (8).^d ND, no data.^e All strains were inducibly clindamycin resistant (Table 2).

addition, both strains were found to harbor an *erm(B)* determinant that differed in several ways from the *erm(B)* determinant described in either the canonical Tn1545 or Tn917 element (15). Strains MSR 63 and MSR610 had a Tn917-like promoter but a 27-amino-acid leader peptide like that found in Tn1545 (Table 3). The two resistant strains also showed a change near the ribosome-binding site (RBS) upstream of the coding sequence. This change may impart better binding of the ribosome and more efficient translation. In addition, these strains had three amino acid differences (I75T, S100N, and H118R) compared to the *erm(B)* coding sequence in Tn1545; however, S100N is found in the *erm(B)* sequence in Tn917

without conferring ketolide resistance. I75T was also present in the *erm(BC)* gene found in the *Escherichia coli* plasmid pIP1527 (3). Only H118R is unique to these ketolide-resistant strains.

emm typing of erythromycin A-resistant ($n = 54$) and randomly selected macrolide-susceptible ($n = 60$) *S. pyogenes* isolates showed that strains of *emm* types 77 ($n = 19$, 45.2%) and 4 ($n = 13$, 26.2%) were found significantly more often among resistant isolates (Table 4).

Antibiotic resistance is of growing concern with regard to *S. pyogenes* isolates from Germany. Previous investigators have reported on macrolide resistance rates among *S. pyogenes* of

TABLE 2. *emm* types and MLST of 54 erythromycin-resistant *S. pyogenes* strains isolated from outpatients in Germany from 1999 to 2000^a

Genotype (<i>n</i>)	MSR strain no.	Town of origin	Source	Infection	MIC (µg/ml)			Macrolide resistance		<i>emm</i>	MLST allele							ST
					ERY	TEL	CLI	GT	PT		<i>gki</i>	<i>gtr</i>	<i>murL</i>	<i>mutS</i>	<i>recP</i>	<i>xpt</i>	<i>yqiL</i>	
<i>mef(A)</i> (30)	18	Dillingen/Saar	Throat swab	Tonsillopharyngitis	1	0.03	0.25	<i>mef(A)</i>	M	1	4	3	4	4	4	2	4	28
	54	Plön	Throat swab	Tonsillopharyngitis	16	0.25	0.25	<i>mef(A)</i>	M	1	4	3	4	4	4	2	4	28
	750	Neukirchen	Swab	Wound infection	16	0.25	0.03	<i>mef(A)</i>	M	1	4	31	2	4	4	2	16	28–V1
	331	Weiden	Throat swab	Tonsillopharyngitis	8	0.5	0.03	<i>mef(A)</i>	M	1	4	3	4	4	4	2V	4	28–V2
	332	Weiden	Throat swab	Tonsillopharyngitis	16	0.5	0.06	<i>mef(A)</i>	M	1	4	3	4	4	4	2V	4	28–V2
	699	Weiden	Throat swab	Tonsillopharyngitis	16	1	0.03	<i>mef(A)</i>	M	1	4	3	4	4	4	2V	4	28–V2
	361	Dillingen/Saar	Throat swab	Tonsillopharyngitis	8	0.5	0.03	<i>mef(A)</i>	M	2	11	9	1	9	2	3	4	55
	576	Dillingen/Saar	Throat swab	Tonsillopharyngitis	2	0.03	0.03	<i>mef(A)</i>	M	3	2	6	8	5	2	3	2	15
	403	Dillingen/Saar	Swab	Wound infection	4	0.125	0.03	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	42	Trier	Throat swab	Tonsillopharyngitis	8	0.25	0.125	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	60	Plön	Throat swab	Tonsillopharyngitis	8	0.25	0.25	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	254	Wülfrath	Throat swab	Tonsillopharyngitis	8	0.125	0.03	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	267	Neukirchen	Throat swab	Tonsillopharyngitis	8	0.25	0.03	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	295	Dillingen/Saar	Throat swab	Tonsillopharyngitis	8	0.25	0.03	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	582	Dillingen/Saar	Ear	Wound infection	8	0.25	0.03	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	598	Plön	Throat swab	Tonsillopharyngitis	8	0.125	0.03	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	51	Plön	Throat swab	Tonsillopharyngitis	16	0.25	0.125	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	52	Plön	Throat swab	Tonsillopharyngitis	16	0.25	0.25	<i>mef(A)</i>	M	4	5	11	8	5	15	2	1	39
	61	Plön	Throat swab	Tonsillopharyngitis	16	0.5	0.06	<i>mef(A)</i>	M	4	4	11	8	5	15	2	1	39–V1
	583	Dillingen/Saar	Throat swab	Tonsillopharyngitis	8	0.25	0.03	<i>mef(A)</i>	M	4	5	11	8	3	15	2	1	39–V2
	5	Leverkusen	Throat swab	Tonsillopharyngitis	16	0.25	0.25	<i>mef(A)</i>	M	4	5	11V	8	5	15	2	1	39–V3
	794	Stuttgart	Throat swab	Tonsillopharyngitis	16	0.25	0.03	<i>mef(A)</i>	M	5	33	30	7	5	5	26	3	99
	260	Wülfrath	Swab	Wound infection	4	0.25	0.03	<i>mef(A)</i>	M	12	5	2	2	6	6	2	2	36
	344	Bonn	Swab	ND	8	0.5	0.06	<i>mef(A)</i>	M	12	5	2	2	6	6	2	2	36
	695	Dillingen/Saar	Throat swab	Tonsillopharyngitis	16	0.25	0.03	<i>mef(A)</i>	M	12	5	2	2	6	6	2	2	36
	773	Neukirchen	Throat swab	Tonsillopharyngitis	16	0.5	0.03	<i>mef(A)</i>	M	12	5	2	2	6	6	2	2	36
	144	Dillingen/Saar	Swab	Wound infection	8	0.25	0.25	<i>mef(A)</i>	M	12	5	11	8	5	15	2	1	39
	591	Bonn	Throat swab	Tonsillopharyngitis	16	0.5	0.03	<i>mef(A)</i>	M	75	11	2	1	3	12	3	7	49
	567	Weiden	Throat swab	Tonsillopharyngitis	2	0.03	0.06	<i>mef(A)</i>	M	77	13	6	2	3	23	3	11	63
	206	Bonn	Ear	ND	16	0.5	0.03	<i>mef(A)</i>	M	77	5	2	2	6	6	2	2	36
<i>erm(B)</i> (7)	564	Weiden	Throat swab	Tonsillopharyngitis	32	0.5	32	<i>erm(B)</i>	c	1	4	3	4	4	4	2V	4	28–V2
	63	Hamburg	Throat swab	Tonsillopharyngitis	32	32	32	<i>erm(B)</i>	c	12	5	2	2	6	6	2	2	36
	235	Bad Hersfeld	Throat swab	Tonsillopharyngitis	32	1	32	<i>erm(B)</i>	c	12	5	2	2	6	6	2	2	36
	610	Stuttgart	Swab	Wound infection	32	4	32	<i>erm(B)</i>	c	22	9	8	1	1	1	3	4	46
	76	Hamburg	Throat swab	Scarlatina	8	0.03	0.25	<i>erm(B)</i>	i	77	13	6	2	3	23	3	11	63
	44	Trier	Throat swab	Tonsillopharyngitis	32	0.03	0.25	<i>erm(B)</i>	i	77	13	6	2	3	23	3	11	63
<i>erm(A)</i> (17)	333	Weiden	Throat swab	Tonsillopharyngitis	32	0.06	0.25	<i>erm(B)</i>	i	77	16	2	8	3	1	13	3	101
	227	Osnabrück	Throat swab	Tonsillopharyngitis	8	0.03	0.125	<i>erm(A)</i>	i	12	3	6	2	3	23	3	11	New
	108	Plön	Throat swab	Tonsillopharyngitis	2	0.03	0.25	<i>erm(A)</i>	i	28	11	6	14	5	9	17	19	52
	247	Trier	Throat swab	Tonsillopharyngitis	2	0.03	0.03	<i>erm(A)</i>	i	28	11	6	14	5	9	17	19	52
	380	Stuttgart	Swab	Wound infection	2	0.03	0.03	<i>erm(A)</i>	i	44/61	4	2	3	11	17	3	1V	New
	600	Wülfrath	Swab	Erysipelas	4	0.03	0.06	<i>erm(A)</i>	i	44/61	4	2	3	1	17	3	1V	New
	393	Leverkusen	Throat swab	Tonsillopharyngitis	4	0.03	0.06	<i>erm(A)</i>	i	58	3	2	3	3	1	3	3	19–V1
	698	Neukirchen	Swab	Wound infection	2	0.03	0.06	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	269	Augsburg	Throat swab	Tonsillopharyngitis	4	0.03	0.125	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	50	Plön	Throat swab	Tonsillopharyngitis	2	0.03	0.03	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	92	Bad Hersfeld	Throat swab	Tonsillopharyngitis	2	0.03	0.25	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	77	Hamburg	Throat swab	Scarlatina	8	0.03	0.125	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	113	Plön	Throat swab	Tonsillopharyngitis	8	0.03	0.25	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	395	Leverkusen	Throat swab	Tonsillopharyngitis	8	0.03	0.125	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	21	Weiden	Swab	Wound infection	32	0.03	0.125	<i>erm(A)</i>	i	77	13	6	2	3	23	3	11	63
	640	Stuttgart	Throat swab	Tonsillopharyngitis	2	0.03	0.03	<i>erm(A)</i>	i	77	13	6	2	3	9	3	11	63–V1
	690	Berlin	Ear	Wound infection	2	0.03	0.03	<i>erm(A)</i>	i	77	9	6	2	5	23	3	11	63–V2
	696	Dillingen/Saar	BS	ND	4	0.03	0.06	<i>erm(A)</i>	i	77	5	6	2	6	23	3	11	63–V3

^a Abbreviations: TEL, telithromycin; ERY, erythromycin; CLI, clindamycin; BS, bronchial secretion; PT, phenotype; GT, genotype; i, inducible MLS_B phenotype; c, constitutive MLS_B phenotype; V, variant; ND, no data.

4% in 1992 to 1993 (5), 1.6% in 1997 (18), 12.7% in 1996 to 1998 (1), 7.9% in 1997 (2), and 13.7% in 2000 (13). In an international study including 1,485 isolates of *S. pyogenes*, almost 10% of *S. pyogenes* isolates were erythromycin A resistant. There was a wide heterogeneity of resistance, with high levels of macrolide resistance in Poland (42%), Hong Kong (28%), Italy (25%), Portugal (24%), and Spain (21%) and no macrolide resistance in Indonesia, Austria, Belgium, The Netherlands, or the United Kingdom (4).

In the present study, stable associations between *emm* type and MLST were observed. Twelve new STs were described. Of note, three strains (MSR 144, MSR 206, and MSR 333) showed an MLST *emm* type combination that had not been described to date. A total of 27 different STs were observed in the present investigation, but three clones were identified to account for more than 50% of macrolide resistance of *S. pyogenes* in Germany: *mef(A)*-positive *emm* type 4 isolates (ST 39, *n* = 10 plus 3 single-locus variants), an *erm(A)*-positive clone

TABLE 3. *erm(B)* upstream sequences from ketolide-resistant *S. pyogenes* isolates

Isolate	Sequence
pAM77	TTTGT--ATAATAGGAATTGAAGTTAA
Tn1545	GT
MSR 63	--
MSR 610	--
pAM77	TTAGATGCTAAAAATTTGTAATTAAGAaggagg
Tn1545	
MSR 63	
MSR 610	
pAM77	GATTCGTCATGTTGGTATTCCAAATGCGTA
Tn1545	
MSR 63	
MSR 610	
pAM77	ATGTAGATAAAACATCTACTGTTTGAACAG
Tn1545	
MSR 63	
MSR 610	
pAM77	ACTAAAAACAGTGATTACGCAGATAAA
Tn1545	
MSR 63	
MSR 610	
pAM77	TA---CGTTAGATTAATTCCTACCAGTGACT
Tn1545	AATA
MSR 63	AATA
MSR 610	AATA
pAM77	AATCTTATGACTTTTTAAACAGATAACTAAAA
Tn1545	
MSR 63	
MSR 610	
pAM77	ATTACAAACAAATCGTTTAACTTCTGTATTTA
Tn1545	
MSR 63	
MSR 610	G
pAM77	TTTATAGATGTA-TCACTTCAGGAGTGATTAC
Tn1545	C A A
MSR 63	A A
MSR 610	A A

^a *erm(B)* upstream sequences from ketolide-resistant *S. pyogenes* isolates are shown. The sequences upstream of *erm(B)* in *Streptococcus sanguis* (pAM77), *S. pneumoniae* (Tn1545), and the two ketolide-resistant *S. pyogenes* strains are compared (15). Boldface residues delineate the promoter region, lowercase letters represent the RBS before a leader peptide (underlined) of 36 amino acids (pAM77) or 27 amino acids (Tn1545, MSR 63, MSR610). (TAA is a stop codon.) Only residues that differ from pAM77 *erm(B)* are shown.

of *emm* type 77 (ST 63, *n* = 8 plus 3 single-locus variants), and *mef(A)*-positive *emm* type 12 isolates (ST 36, *n* = 4).

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TABLE 4. Distribution of *emm* types among 54 erythromycin-resistant and 60 erythromycin-susceptible *S. pyogenes* strains isolated from outpatients in Germany from 1999 to 2000

<i>emm</i> type	No. (%) of strains:	
	Erythromycin susceptible	Erythromycin resistant
1	6 (10.0)	7 (13.0)
2	2 (3.3)	1 (1.9)
3	3 (5.0)	1 (1.9)
4	4 (6.7)	13 (24.1)
5	0 (0.0)	1 (1.9)
6	5 (8.3)	0 (0.0)
9	2 (3.3)	0 (0.0)
12	10 (16.7)	8 (14.8)
22	1 (1.7)	1 (1.9)
28	9 (15.0)	2 (3.7)
29	1 (1.7)	0 (0.0)
44/61	0 (0.0)	2 (3.7)
49	2 (3.3)	0 (0.0)
58	0 (0.0)	1 (1.9)
59	1 (1.7)	0 (0.0)
75	5 (8.3)	1 (1.9)
77	5 (8.3)	16 (29.6)
78	3 (5.0)	0 (0.0)
81	1 (1.7)	0 (0.0)
Total	60 (100.0)	54 (100.0)

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