Letter to the Editor

Molecular Epidemiology of Telithromycin-Resistant Pneumococci in Finland[▽]

In our previous study, we described 26 clinical pneumococci with heterogeneous resistance to telithromycin (TEL). In this resistance type, separate colonies grew inside the inhibition zone around the TEL disk in a disk diffusion test (8). In this study we investigated the molecular epidemiology of these 26 isolates and compared them to *erm*(B)-positive but TEL-susceptible (Tel^s) pneumococci ($n = 66$) (9). Published methods were used for antimicrobial susceptibility testing (7), serotyping (3), pulsed-field gel electrophoresis (PFGE) typing (5, 4), and multilocus sequence typing (MLST) (1). PFGE was performed for all TEL-resistant (Tel^r) isolates, for 3 zone isolates (8), and for 22 randomly selected Tel^s, *erm*(B)-positive isolates, but MLST typing and serotyping were performed only for Tel^r isolates. Our results were compared to international databases (www.mlst.net and www.sph.emory.edu/PMEN).

The clinical characteristics, antimicrobial susceptibilities, PFGE profiles, and sequence types (STs) of the isolates are summarized in Table 1. Penicillin nonsusceptibility (MIC of ≥ 0.125 $\mu\text{g/ml}$) was less frequent among *erm*(B)-positive Tel^r isolates (4/26) than among *erm*(B)-positive Tel^s isolates (50/66) ($P < 0.0001$, Fisher's exact test). The same applied to penicillin and tetracycline coresistance. None of the isolates was resistant to levofloxacin or linezolid. The most frequent serotype was 19A. Tel^r isolates were distributed among 7 distinct PFGE

types, of which type A was the most frequent with 19 isolates. Tel^s *erm*(B) isolates had more heterogeneous PFGE patterns, since 12 different PFGE types were detected among 22 Tel^s isolates (Fig. 1).

MLST types 193, 271, and 273, found among our isolates, are representatives of global pneumococcal clones. ST 193 belongs to the pneumococcal global clone Greece21-30, which has been found in Greece, Brazil, the United Kingdom, and Vietnam (12, 13). The clone was reported to be penicillin susceptible but otherwise multidrug resistant, which also applies to our isolates. ST 273 is a representative of the penicillin-susceptible but erythromycin-resistant PMEN clone, Greece6B-22. Apart from Greece, this clone has been detected in Iceland, Israel, Portugal, Italy, Germany, and Switzerland (6, 11). ST 133 has been detected in Spain (www.mlst.net). ST 271 is a single-locus variant of a multidrug-resistant Taiwanese 19F clone, ST 236 (10). In one international study, ST 271 was detected worldwide and more than 85% of pneumococcal isolates with a double resistance mechanism, *erm*(B) and *mef*(E), belonged to this clonal complex (2). Our ST 271 isolate also had a double resistance mechanism. The other isolate in this study with a double mechanism had a novel ST, ST 2248. Two additional new MLST types were detected: ST 2306 and ST 2307.

TABLE 1. Summary of clinical and molecular data for 26 clinical pneumococcal isolates with heterogeneous resistance to telithromycin

Strain no.	Place of isolation	Patient data			Date of isolation	Susceptibility ($\mu\text{g/ml}$) ^a			Resistance gene(s)	PFGE pattern		Sequence type	Serotype
		Age	Gender	Source		ERY	PEN	TET		Type	Subtype		
115	Helsinki	1	Male	Ear	May 2002	129	0.016	64	<i>erm</i> (B)	B	2	133	18C
897	Lahti	42	Male	BAL ^c	July 2002	129	0.016	64	<i>erm</i> (B)	B	1	133	18C
237	Espoo	2	Male	Ear	May 2002	129	0.016	32	<i>erm</i> (B)	A	3	193	19A
415	Espoo	2	Female	Blood	Oct. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
426	Oulu	32	Female	Sinuitis	Sept. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
427	Oulu	32	Female	Sinuitis	Sept. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
443	Oulu	1	Male	Ear	Sept. 2002	129	0.016	32	<i>erm</i> (B)	A	2	193	19A
460	Oulu	0	Male	Ear	Oct. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
547	Kokkola	2	Female	Sinuitis	Sept. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
552	Kokkola	1	Male	Sinuitis	Oct. 2002	129	0.016	32	<i>erm</i> (B)	A	3	193	19A
683	Kuopio	64	Female	Sputum	May 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
811	Vaasa	78	Female	Blood	May 2002	129	0.016	16	<i>erm</i> (B)	A	3	193	19A
833	Lappeenranta	57	Female	Sinuitis	May 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
859	Lappeenranta	1	Female	BAL	Aug. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
918	Lahti	4	Female	Eye	Sept. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
981	Pori	1	Male	Blood	Aug. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
1016	Pori	0	Male	Blood	Dec. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
1017	Pori	0	Male	Ear	Dec. 2002	129	0.016	32	<i>erm</i> (B)	A	3	193	19A
1022	Pori	1	Male	Ear	Dec. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
1041	Kotka	9	Female	Blood	Oct. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
1051	Kotka	3	Female	Ear	Nov. 2002	129	0.016	32	<i>erm</i> (B)	A	1	193	19A
838	Lappeenranta	1	Female	Ear	May 2002	129	2	32	<i>erm</i> (B), <i>mef</i> (E)	D	1	271	19F
759	Hämeenlinna	2	Female	Ear	Dec. 2002	129	0.016	32	<i>erm</i> (B)	C		273	6B
837	Lappeenranta	8	Male	Sputum	May 2002	129	0.125	64	<i>erm</i> (B), <i>mef</i> (E) ^b	F	1	2248	14
326	Turku	84	Female	Blood	Feb. 2002	129	2	32	<i>erm</i> (B)	G		2306	14
376	Tampere	0	Female	Ear	Oct. 2002	129	2	0.5	<i>erm</i> (B)	E		2307	19F

^a ERY, erythromycin; PEN, penicillin; TET, tetracycline.

^b Isolate 837 also had mutation in L4 (Ser20Asn).

^c BAL, bronchoalveolar lavage.

FIG. 1. Dendrogram and PFGE patterns of 26 *Tel^R*, *erm(B)*-positive pneumococci. *Tel^R* isolates are in boldface. Respective patterns of 22 *erm(B)*-positive but *Tel^S* isolates are also presented. Three isolates designated with the letter V are zone isolates (isolates derived from colonies which grew inside the inhibition zone around the TEL disk in the disk diffusion test) with a pattern identical to those of their parent isolates, 547, 415, and 1022. The dotted line defines PFGE profiles with $\geq 85\%$ similarity (unweighted-pair group method using average linkages, Dice; black vertical dotted line).

The presence of more than one PFGE and MLST type among *Tel^R* isolates suggests that the capability for expressing TEL resistance has arisen several times in *S. pneumoniae*. It is also likely that TEL resistance is an older phenomenon and would not have been caused by TEL selection pressure, because all of our strains were collected in 2002, just before TEL was introduced into clinical practice in Finland. The fact that all of our isolates were of clinical origin means that this type of resistance can survive in the bacterial population and cause diseases to humans. It is also noticeable that six of these isolates were from invasive infections. Our results indicate that pneumococci with heterogeneous TEL resistance can be found in other countries, because many of our strains belonged to global clones. However, this resistance type is difficult to detect using the dilution method (8). We recommend that routine testing of TEL susceptibility of pneumococci be done with the disc diffusion method in a 5%-CO₂ atmosphere, especially in areas where *erm(B)* is a common resistance mechanism.

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Merja Rantala*
Sofia Nyberg
Marianne Lindgren
Pentti Huovinen
Jari Jalava

*Department of Bacterial and Inflammatory Diseases
National Public Health Institute
Kiinamyllynkatu 13
FIN-20520 Turku, Finland*

Reetta Skyttä
Laura Teirilä
Anni Vainio
Anni Virolainen-Julkunen

*Department of Bacterial and Inflammatory Diseases
National Public Health Institute
Helsinki, Finland*

Tarja Kaijalainen
*Laboratory for Chlamydia and Respiratory Tract Bacteria
National Public Health Institute
Oulu, Finland*

*Phone: 358-2-331 6629
Fax: 358-2-331 6699
E-mail: merja.rantala@ktl.fi

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