

Annual survey of methicillin-resistant Staphylococcus aureus (MRSA), 2010

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Introduction

ESR conducts annual surveys of methicillin-resistant *Staphylococcus aureus* (MRSA). Each year, all hospital and community microbiology laboratories in New Zealand are asked to refer all MRSA isolated during a one-month period to ESR. Laboratories provide epidemiological information with each isolate referred. At ESR, MRSA are typed to identify MRSA strains. The purpose of these annual surveys is to provide information on the epidemiology of MRSA in New Zealand and to monitor changes over time.

In 2010, laboratories were asked to refer all MRSA isolated during either August or October 2010 to ESR. The results of the 2010 MRSA survey are presented in this report, along with the trends in MRSA prevalence.

Previous reports on the annual MRSA surveys are available at <u>http://www.surv.esr.cri.nz/antimicrobial/mrsa_annual.php</u>.

Methods

MRSA isolates and data collection

Hospital and community microbiology laboratories in New Zealand were asked to refer all MRSA isolated during a one-month period in 2010 to ESR. LabPlus at Auckland City Hospital, the Microbiology Department at Middlemore Hospital and Medlab South at Nelson Hospital referred MRSA during October 2010 and all remaining laboratories referred MRSA during August 2010. In addition, Whangarei Hospital laboratory reported that they isolated MRSA from diagnostic specimens from eight patients but they did not refer the isolates to ESR. These isolations were included in the analyses of the survey data, except for the analyses relying on MRSA strain identification.

When referring MRSA isolates, laboratories supplied epidemiological data including patient age, geographic location, hospitalisation status, MRSA isolation site, infection or colonisation status, and if MRSA was obtained from a screen or a diagnostic specimen. Laboratories also provided information on the susceptibility of the MRSA isolates to non- β -lactam antibiotics. Two community laboratories in the Auckland area, Labtests and Diagnostic Medlab, receive specimens from multiple district health boards (DHBs), Waitemata, Auckland and Counties Manukau, so these laboratories provided patient or staff addresses that were geocoded at ESR to assign people to a DHB. In 2010, the Otago and Southland DHBs were merged into the Southern DHB. Data from the Otago and Southland DHBs was therefore combined for the years prior to 2010 in order to compare the 2010 data with earlier years and to analyse time trends.

People were classified as hospital patients or hospital staff if (i) they were hospital inpatients or outpatients when MRSA was isolated, or had been in the previous three months; (ii) they were occupying a residential-care facility when MRSA was isolated, or had been in the previous three months; or (iii) they were employed by a healthcare facility (i.e. a hospital or residential-care facility) when MRSA was isolated. Patients or staff were classified as people in the community if (i) MRSA was isolated from patients while in the community and the patients had no history of occupying a healthcare facility in the previous three months; (ii) MRSA was isolated on hospital admission screening of patients who had no history of occupying a healthcare facility in the previous three months; or (iii) MRSA was isolated refacility in the previous three months; or of occupying a healthcare facility in the previous three months; (ii) MRSA was isolated on hospital admission screening of patients who had no history of occupying a healthcare facility in the previous three months; or (iii) MRSA was isolated from pre-employment swabs of healthcare staff with no employment history supplied. All MRSA isolates received at ESR were assumed to be pure cultures of MRSA and methicillin/oxacillin resistance was not routinely confirmed.

spa typing and based upon repeat pattern (BURP) analysis

The polymorphic X region of the staphylococcal protein A gene (*spa*) was amplified as previously described.¹ PCR products were sequenced by the Sequencing Laboratory at ESR using an ABI 3130XL Sequencer. *spa* sequences were analysed using Ridom StaphType software version 2.0.3 (Ridom GmbH, Würzburg, Germany). Sequences were automatically assigned repeats and *spa* types using the software. *spa* types were compared using the BURP algorithm, and by excluding *spa* types with less than five repeats and setting a maximum cost of four between members of a *spa* group cluster.²

Pulsed-field gel electrophoresis (PFGE) and profile analysis

Where necessary to identify strains, PFGE of *Sma*I-digested genomic DNA was performed as previously described.³ PFGE banding patterns were analysed using BioNumerics software version 5.1 (Applied Maths, St-Martens-Latern, Belgium), with the Dice coefficient and unweighted-pair group method with arithmetic averages, at settings of 0.5% optimisation and 1.5% position tolerance. PFGE banding patterns were interpreted using the criteria proposed by Tenover et al.⁴

Multilocus sequence typing (MLST) and sequence analysis

Where necessary to characterise strains, MLST was performed as previously described.⁵ Sequences were analysed using BioNumerics software version 5.1 and sequence types (STs) were assigned using the *S. aureus* database accessible at <u>http://www.mlst.net</u>.

Antibiotic susceptibility testing

Antibiotic susceptibility testing was performed where necessary to identify strains and to supplement the susceptibility information provided by laboratories. Disc susceptibility testing was performed according to the methods of the Clinical and Laboratory Standards Institute (CLSI).⁶ Except for fusidic acid and mupirocin, zones of inhibition were interpreted according to CLSI criteria.⁷ Fusidic acid zones of inhibition were determined with a 10 µg disc and interpreted as \geq 21 mm susceptible, 20 mm intermediate and \leq 19 mm resistant.⁸ Mupirocin zones of inhibition were determined with a 5 µg disc and interpreted as \geq 14 mm susceptible and \leq 13 mm resistant.⁹

PCR for staphylococcal-specific 16S rRNA, nuc and mecA

Isolates that were not able to be *spa* typed were tested for the genes encoding staphylococcal-specific 16S rRNA, *S. aureus*-specific thermostable nuclease (*nuc*) and methicillin resistance (*mecA*) by triplex PCR as previously described.¹⁰

Assigning MRSA strains

Isolates were characterised primarily based upon *spa* types and antibiotic susceptibility patterns, with PFGE as a supplementary typing tool where *spa* typing was inconclusive. There were three situations in which *spa* typing was considered inconclusive: (i) when a *spa* type correlated to a known MRSA strain but the antibiotic susceptibility pattern did not, (ii) when an isolate had a novel *spa* type, and (iii) when an isolate had a *spa* type ESR had not yet correlated to an MRSA strain.

Epidemiological analyses

Epidemiological data and results were entered into ESR's laboratory information management system. Data and results were extracted and analysed using customised Microsoft Access 2003 queries. Point-prevalence rates were calculated based on the number of MRSA isolated per 100 000 population during the period of the survey. The 2001 and 2006 census population data was used to calculate prevalence rates for 2001 and 2006, respectively. For other years, mid-year New Zealand population estimates were used. 95% confidence intervals were calculated based on Poisson distribution. The statistical significance of time trends was calculated at a 95% confidence level using Poisson regression and the Mantel-Haenszel test for linear trend.

Results

National point-prevalence rates of MRSA, 2001-2010

During the 2010 MRSA survey, MRSA were referred or isolated from 754 people, 740 of whom were patients and 14 of whom were staff. There was a 7.5% increase in the MRSA point-prevalence rate between 2009 and 2010, from 16.1 to 17.3 people with MRSA per 100 000 population (Figure 1). Overall, there was a statistically significant (P=0.0067) increase in the MRSA point-prevalence rate over the 10 years, 2001 to 2010 (Figure 1).

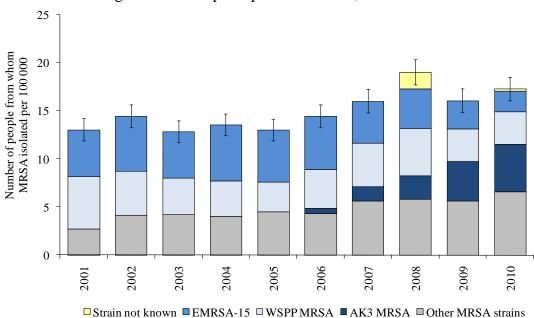


Figure 1. MRSA point-prevalence rates, 2001-2010

95% confidence intervals indicated by error bars. The category 'Strain not known' for 2008 and 2010 represents people identified with MRSA during the survey period but from whom no isolate was referred for strain identification.

MRSA infection status, strain prevalence, and strain association with healthcare facilities versus the community and with patient age

In 2010, of the 740 patients with MRSA, 48.1% were categorised as hospital patients and 51.9% as community patients. MRSA was reported as causing infection in 76.3% of the 653 patients for whom this information was provided.

Six MRSA strains (AK3 MRSA, WSPP MRSA, EMRSA-15, WR/AK1 MRSA, USA300 MRSA and Queensland clone MRSA) were predominant in 2010 and collectively represented 84.8% of all MRSA isolations (Table 1). AK3 MRSA was the most prevalent MRSA strain followed by the WSPP and EMRSA-15 strains. The point-prevalence rates for these three strains were 5.0, 3.4 and 2.2 per 100 000 population, respectively (Figure 1).

Table 1. MRSA strain prevalence, association with healthcare facilities versus the community and association with patient age, 2010

	Proportion (%)	Proportion (%) of each strain isolated from:		
Strain	of all MRSA isolations ^a	hospital patients or staff	people in the community	patients ≥60 years of age ^b
AK3 MRSA	29.0	49.1	50.9	12.4
WSPP MRSA	19.7	24.3	75.7	10.3
EMRSA-15 MRSA	12.5	69.1	30.9	73.3
WR/AK1 MRSA	11.3	54.1	45.9	20.0
USA300 MRSA	7.2	42.6	57.4	18.5
Queensland clone MRSA	5.1	39.5	60.5	5.3

a Other strains accounted for the remaining 15.2% MRSA.

b Age distribution for patients only, staff not included.

Geographic distribution of MRSA

There were geographical differences in the point-prevalence rates of MRSA isolations in 2010, with rates above the national rate of 17.3 MRSA per 100 000 population in the Northland, Auckland, Counties Manukau, Waikato, Lakes, Bay of Plenty and Taranaki DHBs (Figure 2). Similar geographical differences were evident in the point-prevalence rates of MRSA isolated only from infection, however, the rate in the Lakes DHB was below the national point-prevalence rate of 11.4 MRSA infections per 100 000 population, while the rate in Hawke's Bay DHB was above the national average (Figure 3).

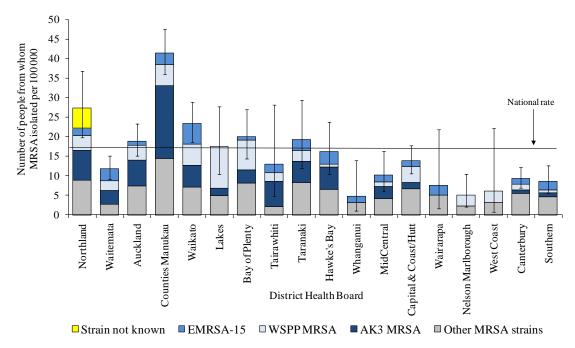


Figure 2. MRSA point-prevalence rates by district health board, 2010

95% confidence intervals indicated by error bars. Data for the Capital & Coast and Hutt DHBs is combined as 'Capital & Coast/Hutt', and data for the Canterbury and South Canterbury DHBs is combined as 'Canterbury'.

There were also geographical differences in the distribution of MRSA strains, particularly AK3 MRSA (Figure 2). The Counties Manukau DHB had the highest point-prevalence rate of this strain, with 18.5 people with AK3 MRSA per 100 000 population (Figure 2) and 9.6 people with AK3 MRSA infection per 100 000 (Figure 3).

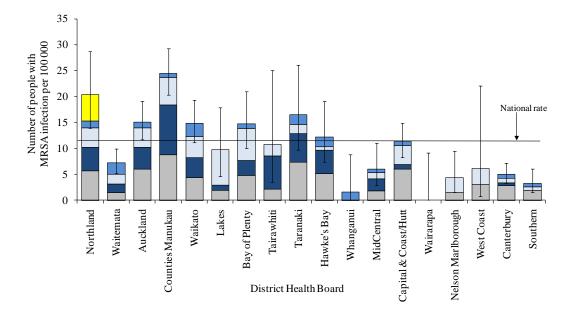


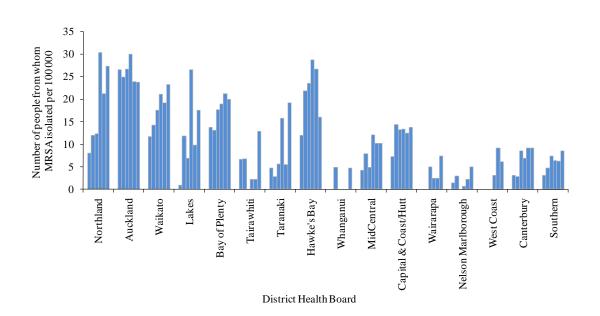
Figure 3. MRSA infection point-prevalence rates by district health board, 2010

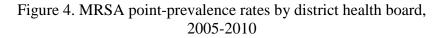
Strain not known infection EMRSA-15 WSPP MRSA AK3 MRSA Other MRSA strains

95% confidence intervals indicated by error bars. Data for the Capital & Coast and Hutt DHBs is combined as 'Capital & Coast/Hutt', and data for the Canterbury and South Canterbury DHBs is combined as 'Canterbury'.

Point-prevalence rates of MRSA by DHB, 2005-2010

Between 2005 and 2010, there were statistically significant increases in MRSA pointprevalence rates in the Northland, Waikato, Lakes, Taranaki and Canterbury DHBs (Figure 4).





The series of bars for each DHB represent the individual years 2005 to 2010 from left to right. Data for the Waitemata, Auckland and Counties Manukau DHBs is combined as 'Auckland', data for the Capital & Coast and Hutt DHBs is combined as 'Capital & Coast/Hutt', and data for the Canterbury and South Canterbury DHBs is combined as 'Canterbury'.

MRSA strain association with spa types

In 2010, the *spa* type most commonly associated with AK3 MRSA was t002 (Table 2). WSPP MRSA was most commonly associated with t019, EMRSA-15 with t032, WR/AK1 MRSA with t127, USA300 MRSA with t008, Queensland clone MRSA with t3949 and AKh4 with t037 (Table 2). EMRSA-15 was associated with the greatest variety of *spa* types (Table 2). There were 109 isolates that were not associated with a known MRSA strain and the most common *spa* types among these isolates were t1853 (17 isolates), t375 (8 isolates), t976 (8 isolates) and t324 (7 isolates). These *spa* types were distinct from each other by BURP analysis and PFGE. The strains associated with these four *spa* types remain to be fully characterised.

	Number		
Strain	of isolates	<i>spa</i> type (number) ^a	spa repeat succession (Ridom)
AK3 MRSA [ST5, SCC <i>mec</i> type IV] ^b	219 ^a	t002 (191)	26-23-17-34-17-20-17-12-17-16
		t306 (5)	26-23-17-34-17-20-17-12-17-17-16
		t548 (4)	26-23-17-34-17-20-17-12-16
		t045 (3)	26-17-20-17-12-17-16
		t062 (3)	26-23-17-12-17-16
		t311 (2)	26-23-17-34-20-17-12-17-16
		t1781 (2)	26-16-16
		t5213 (2)	26-23-17-34-17-20-17-12-12-12-12-16
		t6787 (2)	26-23-17-34-17-20-149-12-17-16
		t242 (1)	26-23-17-13-17-20-17-12-17-16
		t1265 (1)	26-23-17-34-17-20-17-12-12-12-16
		t1340 (1)	26-23-17-34-17-20-17
		t4865 (1)	26-23-17-34-17-13-20-17-12-17-16
		t5867 (1)	26-23-17-36-20-17-12-17-16
WSPP MRSA	148	t019 (138)	08-16-02-16-02-25-17-24
[ST30, SCC <i>mec</i> type IV]		t1347 (3)	08-02-16-02-25-17-24
		t138 (2)	08-16-02-25-17-24
Alternative		t122 (1)	08-16-02-16-02-25-17-24-24
names:		t1836(1)	08-16-02-16-02-25-17-17-24
Southwest Pacific clone and Oceania clone		t2895 (1)	08-16-02-16-02-25-24
		t4341 (1)	08-16-02-17-24
		t4672 (1)	08-16-02-16-02-24-24
EMRSA-15	94	t032 (64)	26-23-23-13-23-31-29-17-31-29-17-25-17-25-16-28
[ST22, SCC <i>mec</i> type IV]		t1401 (7)	26-23-23-13-23-31-29-17-31-29-17-25-17-25-16-28-17-25-16-28
		t022 (6)	26-23-13-23-31-29-17-31-29-17-25-17-25-16-28
		t852 (2)	07-23-13-23-31-05-17-25-17-25-16-28
		t1214 (2)	26-23-23-13-23-31-29-17-31-29-17-25-16-28
		t5501 (2)	26-23-23-13-23-31-29-22-13-23-31-29-17-25-16-28
		t628 (1)	26-23-23-13-23-31-29-17-31-29-17-31-29-17-25-17-25-16-28
		t688 (1)	26-23-17-34-17-16
			continued

Table 2. Frequency of MRSA strains and spa types, 2010

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	Number		
Strain	of isolates	<i>spa</i> type (number)	spa repeat succession (Ridom)
continued EMRSA-15 [ST22, SCCmec type IV]	15014105	t718 (1)	26-23-23-13-23-31-29-17-31-29-17-25-17-25-16-28
		t788 (1)	26-23-23-13-23-24-25-17-25-16-28
		t906 (1)	07-23-31-29-17-31-29-17-25-17-25-16-28
		t1415 (1)	26-23-23-13-23-31-29-17-31-29-16-28
		t1467 (1)	26-16-23-31-29-17-31-29-17-25-17-25-16-28
		t5538 (1)	26-23-23-20-13-23-31-29-17-31-29-17-25-17-25-16-28-17-25-16-28
		t6448 (1)	26-23-23-31-29-17-31-29-17-25-17-25-16-75-28
		t6525 (1)	26-23-23-13-23-31-29-17-25
		t7184 (1)	07-29-17-31-29-17-25-17-25-16-28
WR/AK1 MRSA	84 ^c	t127 (73)	07-23-21-16-34-33-13
[ST1, SCCmec		t701 (6)	11-10-21-17-34-24-34-22-25-25
type IV]		t177 (1)	26-23-21-16-34-33-13
A 14		t267 (1)	07-23-12-21-17-34-34-33-34
Alternative name: Western		t359 (1)	07-23-12-21-17-34-34-33-34
Australia (WA)		t693 (1)	07
MRSA-1		t7136(1)	07-23-12-21-21-17-34-34-33-34
USA300 MRSA	54	t008 (45)	11-19-12-21-17-34-24-34-22-25
[ST8, SCC <i>mec</i> type IV]		t024 (4)	11-12-21-17-34-24-34-22-25
type Iv]		t1882 (2)	11-19-12-21-22-17-34-24-34-22-25
		t1627 (1)	11-10-12-21-17-34-24-34-22-25
		t2558 (1)	11-19-12-12-34-22-25
		t5498 (1)	11-12-21-17-34-24-21-25
Queensland	38	t3949 (27)	11-17-23-17-17-16-16-25
clone MRSA [ST93, SCC <i>mec</i> type IV]		t202 (9)	11-17-23-17-17-16-16-25
		t6487 (1)	11-17-23-17-17-16-16
		t7238 (1)	11-17-23-17-16-25
AKh4 MRSA [ST239, SCCmec type III]	5	t037 (3)	15-12-16-02-25-17-24
		t631 (1)	15-12-16-17
		t4866 (1)	15-12-16-12-16-02-25-17-24
Alternative names: EMRSA-1, AUS-2 EMRSA and AUS-3 EMRSA			

Table 3. Frequency of MRSA strains and spa types, 2010 continued

a One person had two different *spa* types, t002 and t6787, both were the AK3 MRSA strain.
b ST, multilocus sequence type; SCC*mec*, staphylococcal cassette chromosome *mec*.
c Total number of WR/AK1 MRSA isolates was 85 but one isolate was not typable by *spa* typing.

Discussion

The prevalence of MRSA in New Zealand continues to rise, with the point-prevalence rate significantly increasing during the 10 years, 2001-2010, and increasing by 7.5% from 2009 to 2010. This increase is reflected particularly in the Northland, Waikato, Lakes, Taranaki and Canterbury DHBs, which all demonstrated significant increases in MRSA point-prevalence rates over the six years, 2005-2010.

Consistent with earlier years, in 2010 there were large geographical differences in the prevalence of MRSA within New Zealand, with rates generally highest in DHBs in the upper half of the North Island. As MRSA from both diagnostic specimens and screening specimens were included in the survey, any apparent differences in MRSA rates between DHBs could be partly due to differences in screening policies. However, the relative rates of MRSA infections between DHBs were very similar to the rates of all MRSA isolations. Rates of MRSA infections may also be influenced by different policies for obtaining and processing diagnostic specimens.

Eight MRSA strains are currently recognised in New Zealand: AK3 MRSA [ST5, SCC*mec* type IV], AKh4 MRSA [ST239, SCC*mec* type III], EMRSA-15 [ST22, SCC*mec* type IV], EMRSA-16 [ST36, SCC*mec* type II], Queensland clone MRSA [ST93, SCC*mec* type IV], USA300 MRSA [ST8, SCC*mec* type IV], WR/AK1 MRSA [ST1, SCC*mec* type IV] and WSPP MRSA [ST30, SCC*mec* type IV]. Supplementary descriptions of these strains, including typical antibiotic susceptibility patterns, are available at <u>http://www.esr.cri.nz/competencies/Health/Pages/MRSA% 20strains.aspx</u>. In 2010, AK3 MRSA, WSPP MRSA, EMRSA-15, WR/AK1 MRSA, USA300 MRSA, and Queensland clone MRSA were collectively responsible for 84.8% of MRSA isolations in New Zealand.

Each year since 2005, the prevalence of AK3 MRSA has increased, and in 2010 this strain accounted for the highest proportion (29.0%) of MRSA isolations. AK3 MRSA was most prevalent in the Counties Manukau DHB followed by other DHBs in the upper half of the North Island. AK3 MRSA has yet to become as widespread as the previously predominant community-associated MRSA in New Zealand – WSPP MRSA, which is still widely distributed throughout the country.

In 2010 the overall distribution of MRSA between hospital patients or staff and people in the community was consistent with 2009.¹¹ As in previous years, WSPP MRSA was mainly associated with people in the community, EMRSA-15 was mainly associated with hospital patients or staff, and USA300 MRSA was isolated in similar proportions from hospital patients or staff and people in the community.¹¹ Consistent with reports from Australia, where the Queensland clone is primarily community-associated, 60.5% of Queensland clone MRSA isolates were from people in the community.¹²

Although AK3 MRSA and WR/AK1 MRSA are considered to be communityassociated, 49.1% of AK3 MRSA isolates and 54.1% of WR/AK1 MRSA included in the 2010 survey were from hospital patients or staff. However, the epidemiological information we are able to collect on people with MRSA only allows us to categorise where a person was when their MRSA was isolated or if they had been in a healthcare facility in the last 3 months. We do not attempt to categorise people according to where they acquired their MRSA. Therefore, it is likely that some people who have acquired MRSA in the community will be categorised as 'hospital patients or staff', and conversely, that some people who have acquired MRSA in a healthcare facility will be categorised as 'people in the community'. However, the young age profile of the patients with AK3 MRSA and WR/AK1 MRSA is typical of community-associated MRSA, with only 12.4% and 20.0%, respectively, being isolated from patients \geq 60 years of age.¹³ Moreover, these two strains are not usually multiresistant – another feature typical of community-associated MRSA.

References

- 1 Strommenger B, Braulke C, Heuck D, Schmidt C, Pasemann B, Nübel U, et al. *spa* typing of *Staphylococcus aureus* as a frontline tool in epidemiological typing. J Clin Microbiol 2008; 46: 574-81.
- 2 Mellmann A, Weniger T, Berssenbrugge C, Rothganger J, Sammeth M, Stoye J, et al. Based upon repeat pattern (BURP): an algorithm to characterize the long-term evolution of *Staphylococcus aureus* populations based on spa polymorphisms. BMC Microbiol 2007; 7: 98.
- 3 Goering RV. Pulsed-field gel electrophoresis. In: Persing DH, Tenover FC, Versalovic J, Tang YW, Unger ER, Relman DA, White TJ, editors. Molecular microbiology: diagnostic principles and practice. Washington: ASM Press; 2004. p. 185-96.
- 4 Tenover FC, Arbeit RD, Goering RV, Mickelsen PA, Murray BE, Persing DH, et al. Interpreting chromosomal DNA restriction patterns produced by pulsed- field gel electrophoresis: criteria for bacterial strain typing. J Clin Microbiol 1995; 33: 2233-9.
- 5 Enright MC, Day NP, Davies CE, Peacock SJ, Spratt BG. Multilocus sequence typing for characterization of methicillin-resistant and methicillin-susceptible clones of *Staphylococcus aureus*. J Clin Microbiol 2000; 38: 1008-15.
- 6 Clinical and Laboratory Standards Institute. Performance standards for antimicrobial disk susceptibility tests; approved standard – tenth edition. Wayne (PA): CLSI; 2009. CLSI document M2-A10.
- 7 Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing; twentieth informational supplement. Wayne (PA): CLSI; 2010. CLSI document M100-S20.
- 8 Toma E, Barriault D. Antimicrobial activity of fusidic acid and disk diffusion susceptibility testing criteria for gram-positive cocci. J Clin Microbiol 1995; 33: 1712-5.
- 9 Finlay JE, Miller LA, Poupard JA. Interpretive criteria for testing susceptibility of staphylococci to mupirocin. Antimicrob Agents and Chemother 1997; 41: 1137-9.
- 10 Maes N, Magdalena J, Rottiers S, De Gheldre Y, Struelens MJ. Evaluation of a triplex PCR assay to discriminate *Staphylococcus aureus* from coagulase-negative Staphylococci and determine methicillin resistance from blood cultures. J Clin Microbiol 2002; 40: 1514-7.
- 11 Richardson A, Pope C, Couper J, Desai U, Heffernan H. Annual survey of methicillin-resistant *Staphylococcus aureus* (MRSA), 2009. Porirua: Institute of Environmental Science and Research; 2010. Available at <u>http://www.surv.esr.cri.nz/PDF_surveillance/Antimicrobial/MRSA/aMRSA_2008</u>. <u>pdf</u>.

- 12 Coombs G, Pearson J, Christiansen K, Nimmo G, on behalf of the Australian Group for Antimicrobial Resistance (AGAR). *Staphylococcus aureus* programme 2008 (SAP 2008) community survey MRSA epidemiology and typing report; 2009. Available at <u>http://www.agargroup.org/files/FED% 20REPORT% 20SAP2008% 20MRSA% 20final.pdf</u>.
- 13 Naimi TS, LeDell KH, Como-Sabetti K, Borchardt SM, Boxrud DJ, Etienne J, et al. Comparison of community- and health care-associated methicillin-resistant *Staphylococcus aureus* infection. JAMA 2003; 22: 2976-84.