Community-onset *Staphylococcus aureus* Surveillance Programme annual report, 2012

Geoffrey W Coombs, Denise A Daley, Julie C Pearson, Graeme R Nimmo, Peter J Collignon, Mary-Louise McLaws, James O Robinson, John D Turnidge for the Australian Group on Antimicrobial Resistance

Abstract

In 2012, the Australian Group on Antimicrobial Resistance (AGAR) conducted a community-onset period-prevalence survey of clinical *Staphylococcus aureus* isolated from hospital outpatients and general practice patients including nursing homes, long term care facilities and hospice patients. Day surgery and dialysis patients were excluded. Twenty-nine medical microbiology laboratories from all state and mainland territories participated. Isolates were tested by Vitek2® (AST-P612 card). Results were compared with previous AGAR community surveys. Nationally, the proportion of *S. aureus* that were methicillin-resistant *S. aureus* (MRSA) increased significantly from 11.5% in 2000 to 17.9% in 2012 ($P<0.0001$). Resistance to the non-ß-lactam antimicrobials varied between regions. No resistance was detected to vancomycin, teicoplanin or linezolid. Resistance in methicillin susceptible *S. aureus* was rare apart from erythromycin (12.8%) and was absent for vancomycin, teicoplanin, linezolid and daptomycin. The proportion of *S. aureus* characterised as health care-associated MRSA (HA-MRSA) was 5.1%. Three HA-MRSA clones were characterised, with 72.9% and 26.4% of HA-MRSA classified as ST22-IV [2B] (EMRSA-15) and ST239-III [3A] (Aus-2/3 EMRSA) respectively. Multi-clonal community-associated MRSA (CA-MRSA) accounted for 12.5% of all *S. aureus*. Regional variation in resistance in MRSA was primarily due to the differential distribution of the 2 major HA-MRSA clones; ST239-III [3A] (Aus-2/3 EMRSA), which is resistant to multiple non-ß-lactam antimicrobials, and ST22-IV [2B] (EMRSA-15), which is resistant to ciprofloxacin and typically erythromycin. Although the majority of CA-MRSA were non-multi-resistant, a significant expansion of Panton-Valentine leukocidin (PVL) positive CA-MRSA clones has occurred nationally. The mean age of patients (31.7 years, 95% CI 28.9–34.5) with a PVL positive CA-MRSA infection was significantly lower ($P<0.0001$), than the mean age of patients with a PVL negative CA-MRSA infection (55.7 years, 95% CI 50.7–60.6). This shift in the molecular epidemiology of MRSA clones in the Australian community will potentially increase the number of young Australians with skin and soft tissue infections requiring hospitalisation. *Commun Dis Intell* 2014;38(1):E59–E69.

Keywords: antimicrobial resistance surveillance; *Staphylococcus aureus;* community-onset; methicillin resistance

Introduction

Staphylococcus aureus continues to be the causative organism of a wide range of community-acquired infections ranging from relatively minor skin and soft tissue infections to serious and life threatening systemic sepsis with a high mortality.[1,2] In Australia, methicillin-resistant *S. aureus* (MRSA) was first detected in Sydney in the 1960s,[3] but really became an endemic problem in hospitals, in particular in the eastern states, with the appearance of a multi-resistant strain, (Aus-2/3 EMRSA), in the 1970s and 80s.[4,5] In Australia, community-associated MRSA (CA-MRSA) strains emerged in the 1990s, initially in Western Australia and the Northern Territory,[6–8] and subsequently in the eastern states.[9–11] These MRSA strains are generally less resistant to a range of antimicrobials and associated with skin and soft tissue infection (SSTI). Strains harbouring the genes encoding Panton-Valentine leukocidin (PVL) were first detected in Australia in the late 1990s (the South Western Pacific [SWP] or Oceania clone: ST30-IV [2B].[12] The PVL positive Queensland clone (ST93-IV [2B]) was characterised in 2000 and is now the dominant CA-MRSA in Australia.[13,14] Importation of several overseas PVL positive clones has occurred: USA300 (ST8-IV [2B]), the Bengal Bay Clone (ST772-V [5C2]), Taiwan CA-MRSA (ST59-V [5C2 and 5]) and European CA-MRSA (ST80-IV [2B]).[15] PVL is associated with recurrent furunculosis and more severe infections including osteomyelitis, septicaemia and necrotising pneumonia.

The Australian Group on Antimicrobial Resistance (AGAR) has conducted surveillance of antimicrobial resistance in *S. aureus* for over 20 years.[16] This surveillance role is very important given the ability of *S. aureus* strains to acquire new resistance and virulence determinants and to undergo rapid clonal expansion. Since the 1960s multiple waves of MRSA clones have occurred in Australia influencing the susceptibility profiles of the isolates seen in clinical practice. Results of previous AGAR surveys provide the only longitudinal record of the epidemiology of MRSA at a national level.[17–19] Given the emergence

of hyper-virulent community MRSA strains, AGAR changed its methodology in 2000 to conduct surveys of community isolates biennially. The community-based surveys performed in 2000, 2002, 2004 and 2006 have been reported previously.[20–22]

The results of the 7th community-based survey of *S. aureus* infection conducted in 2012 are reported here.

Methods

Twenty-nine laboratories from all 8 Australian states and territories participated in the 2012 *S. aureus* AGAR survey.

From 1 July to 30 November 2012 each laboratory collected up to 100 clinically significant consecutive *S. aureus* isolates from different patients. Isolates were collected from hospital outpatients. Day surgery and dialysis patients were excluded. Isolates from nursing homes, long-term care facilities and hospice patients were included. Each *S. aureus* isolate was from an individual patient and was judged to have come from a potentially infected site.

Susceptibility methodology

All isolates were tested using the Vitek2® AST-P612 card. All isolates with a penicillin minimum inhibitory concentration (MIC) of ≤0.125 mg/L were screened for the presence of ß-lactamase using nitrocefin or disc diffusion using a Penicillin 10 unit disc (CLSI) or Penicillin 1 unit disc (EUCAST). High-level mupirocin resistance was determined by disc diffusion (200 ug). CLSI breakpoints were utilised for all antimicrobials[23] except fusidic acid (http://www.eucast.org/clinical_breakpoints/). Isolates with an MIC in the intermediate resistance category have been called resistant in this report.

Epidemiological typing of methicillin-resistant *Staphylococcus aureus*

Of the 510 MRSA identified, 499 (97.8%) were referred to the Australian Collaborating Centre for Enterococcus and Staphylococcus Species (ACCESS) Typing and Research for epidemiological typing.

Electrophoresis of chromosomal DNA using a contour-clamped homogeneous electric field DRIII System (Bio-Rad Laboratories Pty Ltd) was performed as previously described on all MRSA isolates.[24] Multilocus sequence typing (MLST) and SCC*mec* typing was performed as previously described on selected MRSA isolates.[25–27]

PCR for the detection of PVL determinants was performed as previously described on all MRSA isolates.[28]

Methicillin-resistant *Staphylococcus aureus* nomenclature

MRSA clones were defined by the combination of the multilocus sequence type (ST) and the SCC*mec* type.[29] Clones are reported with their ST and SCC*mec* type followed by their colloquial name in parenthesis; e.g. ST22-IV [2B] (EMRSA-15). SCC*mec* nomenclature is used as proposed by the International Working Group on the Classification of Staphylococcal Cassette Chromosome Elements.[30] Briefly, the structural type is indicated by a Roman numeral, with a lowercase letter indicating the subtype, and the *ccr* complex and the *mec* complex are indicated by an Arabic numeral and an uppercase letter respectively in parenthesis. Where there is an extra *ccr* element, this is indicated by '&' and an Arabic numeral designating the *ccr* type. When there is an extra *ccr* element present whose precise location is unknown it is indicated by an '&' and *ccr* number outside the parentheses. Clones were classified into two groups on the basis of previously published evidence: those implicated in healthcare-associated infection (HA-MRSA) and those implicated in CA-MRSA.

Clones that diverged at no more than one of the 7 MLST loci were considered to belong to the same clonal complex (CC). Double locus variants were included in the same CC if the linking single locus variant was present in the MLST database (http://www.mlst.net/).

Statistical analysis

The difference between proportions was tested using a Chi-square test and Fisher's exact test (GraphPad® Prism Software). Relative risk and 95% confidence intervals (CI) were calculated using VassarStats (http://vassarstats.net).

Results

The survey included 2,844 isolates (Table 1) with the majority (1,792, 63.0%) being contributed by New South Wales, Queensland and Victoria.

SSTI specimens contributed the majority of isolates (2,575, 90.5%) followed by respiratory specimens (106, 3.7%) and bacteraemia (89, 3.1%). There were significantly ($P<0.0001$) more isolates causing non-invasive (2,740, 96.3%) than invasive (104, 3.7%) infections (Table 2).

Table 1: *Staphylococcus aureus* isolates, Australia, 2012, by state or territory

State or territory	Number of institutions	Total	%
ACT	1	100	3.5
NSW	7	693	24.4
NT	1	100	3.5
Qld	6	599	21.1
SA	3	296	10.4
Tas.	2	159	5.6
Vic.	5	500	17.6
WA	4	397	14.0
Total	29	2,844	100.0

The proportion of *S. aureus* that were MRSA was 17.9% (95% CI 16.6–19.4%) nationally (Table 3), ranging from 4.0% in the Australian Capital Territory to 25.5% in New South Wales.

Table 2: Source of *Staphylococcus aureus* isolates, Australia, 2012

Specimen source	Number	%	95% CI
Skin and soft tissue	2,575	90.5	89.4–91.6
Respiratory	106	3.7	3.1–4.5
Blood	89	3.1	2.6–3.8
Urine	58	2.0	1.6–2.6
Sterile body cavity	16	0.6	0.4–0.9
Total	2,844		
Invasive*	104	3.7	3.0–4.4
Non-invasive	2,740	96.3	95.6–97.0

* Blood or sterile body cavity

The proportion of invasive isolates (blood/sterile body cavity sites) that were MRSA was 21.2% (95% CI 14.4–30.0%) and was similar (P=0.4037) to the proportion of non-invasive isolates at 17.8% (95% CI 16.4–19.3%) (Table 3). The proportion of MRSA was highest in blood at 21.3% (95% CI 14.1–31.0%) (Table 4).

There were significant differences (P<0.0001) in the proportion of MRSA seen in different patient groups with patients from long term care facilities (46.7%, 95% CI 24.8–70.0%), patients attending emergency departments (20.9%, 95% CI 18.9–23.1%) and hospital outpatients (17.0%, 95% CI 14.2–20.1%) having high rates of MRSA. In general practice patients the proportion of *S. aureus* that were MRSA was 12.7% (95% CI 10.5–15.3%).

Resistance in MRSA to non-ß-lactam antimicrobials (with the exception of rifampicin, high-level mupirocin and fusidic acid) varied between regions (Table 5). Two isolates were non-susceptible to daptomycin. No resistance was detected to vancomycin, teicoplanin or linezolid. There were differences in the proportion of isolates resistant to non-ß-lactam antimicrobials in MRSA associated with

Table 4: Proportion of *Staphylococcus aureus* that were methicillin resistant, Australia, 2012, by specimen type

Source of infection	n/N	%	95% CI
Blood	19/89	21.3	14.1–31.0
Sterile body cavity	3/16	18.8	6.6–43.1
Skin and soft tissue	462/2,575	17.9	16.5–19.5
Urine	10/58	17.2	9.6–28.9
Respiratory	16/106	15.1	9.5–23.1
Total	510/2,844	17.9	16.6–19.4

Table 3: Proportion of *Staphylococcus aureus* that were methicillin resistant, Australia, 2012, by state or territory and source

State or territory	All isolates			Invasive isolates*			Non-invasive isolates		
	n/N	%	95% CI	n/N	%	95% CI	n/N	%	95% CI
ACT	4/100	4.0	1.2–10.2	0/0	0.0		4/100	4.0	1.2–10.2
NSW	177/693	25.5	22.4–28.9	13/33	39.4	24.7–56.4	164/660	24.8	21.7–28.3
NT	24/100	24.0	16.6–33.3	1/2	50.0	9.5–90.6	23/98	23.5	16.1–32.8
Qld	103/599	17.2	14.4–20.4	2/17	11.8	2.0–35.6	101/582	17.4	14.5–20.7
SA	43/296	14.5	10.9–19.0	1/5	20.0	2.0–64.0	42/291	14.4	10.8–19.0
Tas.	9/159	5.7	2.9–10.6	0/11	0.0	0–30.0	9/148	6.1	3.1–11.3
Vic.	87/500	17.4	14.3–21.0	3/23	13.0	3.7–33.0	84/477	17.6	14.4–21.3
WA	63/397	15.9	12.6–19.8	2/13	15.4	3.1–43.5	61/384	15.9	12.6–19.0
Aus.	510/2,844	17.9	16.6–19.4	22/104	21.2	14.4–30.0	488/2,740	17.8	16.4–19.3

* Blood/sterile body cavity

Table 5: Number and proportion of methicillin-resistant *Staphylococcus aureus* resistant to the non-ß-lactam antimicrobials, Australia, 2012, by state or territory

	ACT (n=4)		NSW (n=177)		NT (n=24)		Qld (n=103)		SA (n=43)		Tas. (n=9)		Vic. (n=87)		WA (n=63)		Aus. (n=510)		Differences across regions	
Drug	n	%	n	%	n	%	n	%	N	%	n	%	n	%	n	%	n	%	X^2	*P*
Erythromycin	3	75.0	77	43.5	7	29.2	22	21.4	14	30.2	4	44.4	51	58.6	23	36.5	200	39.2	31.05	<0.0001
Clindamycin*	0	0.0	28	15.8	3	12.5	10	9.7	5	11.6	0	0.0	20	23.0	2	3.2	68	13.3	16.89	0.0182
Tetracycline	0	0.0	28	15.8	5	20.8	9	8.7	5	11.6	0	0.0	26	29.9	1	1.6	74	14.5	31.34	<0.0001
Co-trimoxazole	0	0.0	20	11.3	3	12.5	4	3.9	3	7.0	0	0.0	19	21.8	3	4.8	52	10.2	21.73	0.0028
Ciprofloxacin	1	25.0	91	51.4	4	16.7	20	19.4	8	18.6	4	44.4	51	58.6	13	20.6	191	37.5	64.53	<0.0001
Gentamicin	0	0.0	22	12.4	5	20.8	3	2.9	2	4.7	0	0.0	14	16.1	2	3.2	48	9.4	20.59	0.0044
Fusidic acid	0	0.0	8	4.5	0	0.0	7	6.8	2	4.7	0	0.0	7	8.0	2	3.2	26	5.1	4.786	0.6861
Mupirocin†	0	0.0	4	2.3	0	0.0	4	3.9	0	0.0	0	0.0	0	0.0	0	0.0	8	1.6	7.788	0.3517
Rifampicin	0	0.0	2	1.1	0	0.0	1	1.0	0	0.0	0	0.0	2	2.3	1	1.6	6	1.2	2.027	0.9583

* Constitutive resistance.

† High-level resistance.

Table 6: Number and proportion of methicillin sensitive *Staphylococcus aureus* resistant to the non-ß-lactam antimicrobials, Australia, 2012, by state or territory

	ACT (n=96)		NSW (n=516)		NT (n=76)		Qld (n=496)		SA (n=253)		Tas. (n=150)		Vic. (n=413)		WA (n=334)		Aus. (n=2,334)		Differences across regions	
Drug	n	%	n	%	n	%	n	%	N	%	n	%	n	%	n	%	n	%	X^2	*P*
Penicillin	81	84.4	451	87.4	69	90.8	436	87.9	213	84.2	125	83.3	363	87.9	275	82.3	2,013	86.2	10.56	0.1589
Erythromycin	14	14.6	64	12.4	12	15.8	61	12.3	39	15.4	18	12.0	59	14.3	32	9.6	299	12.8	6.616	0.47
Clindamycin*	1	1.0	12	2.3	0	0	8	1.6	5	2.0	1	0.7	7	1.7	4	1.2	38	1.6	4.487	0.7223
Tetracycline	5	5.2	14	2.7	0	0.0	8	1.6	8	3.2	5	3.3	17	4.1	16	4.8	73	3.1	12.28	0.0918
Co-trimoxazole	5	5.2	21	4.1	0	0.0	5	1.0	12	4.7	3	2.0	24	5.8	7	2.1	87	3.7	24.94	0.0008
Ciprofloxacin	2	2.1	11	2.1	1	1.3	13	2.6	8	3.2	8	5.3	21	5.1	9	2.7	65	2.8	11.12	0.1337
Gentamicin	1	1.0	3	0.6	0	0.0	3	0.6	3	1.2	0	0.0	3	0.7	0	0.0	13	0.6	5.590	0.5884
Fusidic acid	7	7.3	33	6.4	2	2.6	56	11.3	3	1.2	10	6.7	13	3.1	11	3.3	135	5.8	48.84	<0.0001
Mupirocin†	2	2.1	8	1.6	1	1.3	47	9.5	2	0.8	0	0.0	4	1.0	4	1.2	68	2.9	97.36	<0.0001
Rifampicin	0	0.0	3	0.6	0	0.0	2	0.4	0	0.0	0	0.0	2	0.5	0	0.0	7	0.3	4.748	0.6907

* Constitutive resistance.

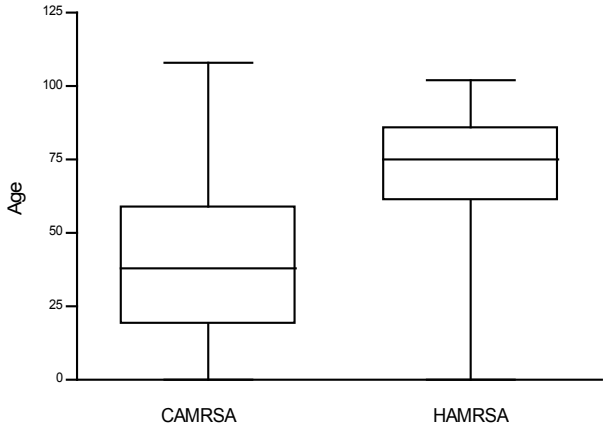
† High-level resistance.

various patient types. MRSA resistance for many antimicrobials was high in hospital outpatients, emergency and long-term care, which is consistent with a higher proportion of these having been acquired in healthcare-related settings.

Susceptibility testing of methicillin sensitive *S. aureus* (MSSA) (Table 6) show resistance to non-ß-lactam antimicrobials remains uncommon except for erythromycin where overall resistance was 12.8% (95% CI 11.5–14.2%). All isolates were susceptible to vancomycin, teicoplanin, linezolid and daptomycin. Resistance to penicillin was high and in similar proportions ranging from 82.3% to 90.8% across all regions.

Based on molecular typing, of the 499 MRSA referred to ACCESS Typing and Research, 28.9% (144) and 71.1% (355) were classified as HA-MRSA and CA-MRSA strains respectively. The mean age of patients with a CA-MRSA infection (40.6 years, 95% CI 37.8–43.4) was significantly lower ($P<0.0001$), than the mean age of patients with a HA-MRSA infection (69.8 years 95% CI 66.2–73.4) (Figure 1).

Figure 1: Box plot of age of patients infected with community-associated methicillin-resistant *Staphylococcus aureus* and healthcare-associated methicillin-resistant *Staphylococcus aureus*, Australia, 2012



Community-associated methicillin-resistant *Staphylococcus aureus* Mean age 40.6 years (95% CI: 37.8–43.4).

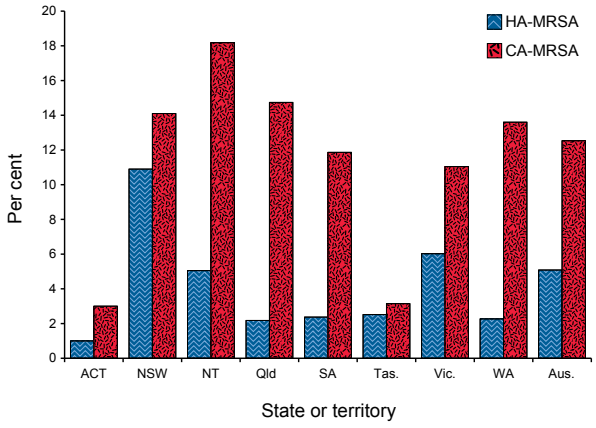
Healthcare-associated methicillin-resistant *Staphylococcus aureus* Mean age 69.8 years (95% CI: 66.2–73.4).

Throughout Australia, the percentage of *S. aureus* characterised as HA-MRSA was 5.1%, ranging from 1.0% in the Australian Capital Territory to 10.8% in New South Wales (Figure 1). Three HA-MRSA clones were identified: ST22-IV [2B] (EMRSA-15) (72.9% of HA-MRSA), ST239-III [3A] (Aus-2/3 EMRSA) (26.4%),and 1 isolate of ST5-II [2A] (New York Japan MRSA/USA100).

ST22-IV [2B] (EMRSA-15) has become the predominant HA-MRSA clone in the Australian community accounting for 21.0% of MRSA ranging from 0% in the Northern Territory to 44.4% in Tasmania (Table 7). Typically PVL negative, 99% and 61% of ST22-IV [2B] (EMRSA-15) isolates were resistant to ciprofloxacin and erythromycin respectively.

ST239-III [3A] (Aus-2/3 EMRSA) accounted for 7.6% of MRSA ranging from 0% in the Australian Capital Territory and Tasmania to 21.7% in the Northern Territory (Table 7). PVL negative ST239-III [3A] (Aus-2/3 EMRSA was typically resistant to tetracycline (100%), erythromycin (100%), gentamicin (97%),ciprofloxacin (92%), and cotrimoxazole (92%).

Figure 2: Percentage of *Staphylococcus aureus* characterised as healthcare-associated methicillin-resistant *Staphylococcus aureus* and community-associated methicillin-resistant *Staphylococcus aureus* strains, Australia, 2012, by state or territory



Throughout Australia, the percentage of *S. aureus* characterised as CA-MRSA was 12.5%, ranging from 3.0% in the Australian Capital Territory to 18.0% in the Northern Territory (Figure 2). Thirty-two CA-MRSA clones were identified by pulsed-field gel electrophoresis, corresponding to 25 MLST/SCC*mec* clones (Table 8). Overall, 82.5% of CA-MRSA were classified into 6 clones: ST93-IV [2B] (Qld CA-MRSA) (36.3% of CA-MRSA);ST30-IV [2B] (SWP MRSA) (16.9%); ST1-IV [2B] (WA1) (13.5%); ST45-V [5C2&5] (WA84) (5.9%); ST78-IV [2B] (WA2) (5.1%); and ST5-IV [2B] (WA3) (4.8%).

Table 7: Proportion of methicillin-resistant *Staphylococcus aureus* characterised as ST22-IV [2B] (EMRSA-15) and ST239-III [3A] (Aus-2/3 EMRSA), Australia, 2012, by state or territory

	ACT	`NSW	NT	Qld	SA	Tas.	Vic.	WA	Aus.
ST22-IV [2B]	25.0	34.3	0.0	9.9	11.9	44.4	21.2	12.7	21.0
ST239-III [3A]	0.0	8.7	21.7	3.0	4.8	0.0	14.1	1.6	7.6

Table 8: Proportion of community-associated methicillin-resistant *Staphylococcus aureus*, Australia, 2012, by clone and Panton Valentine leukocidin carriage

Clone	Clonal complex	Alternative name	n (%)	PVL pos (%)
ST93-IV [2B]	Singleton	Queensland MRSA	129 (36.3)	127 (98.4)
ST30-IV [2B]	30	SWP MRSA	60 (16.9)	56 (93.3)
ST1-IV [2B]	1	WA1	48 (13.5)	3 (6.3)
ST45-V [5C2&5]	45	WA84 (Vic CA-MRSA)	21 (5.9)	0
ST78-IV [2B]	88	WA2	18 (5.1)	1 (5.6)
ST5-IV [2B]	5	WA3	17 (4.8)	5 (29.4)
ST73-IV [2B]	5	WA65	10 (2.8)	0
ST8-IV [2B]	8	USA300	10 (2.8)	9 (90.0)
ST952-V [5C2&5]	59	Taiwan A MRSA	5 (1.4)	5 (100)
ST59-V [5C2&5]	59	Taiwan MRSA	5 (1.4)	5 (100)
ST5-IV [2B]	5	WA121	4 (1.1)	4 (100)
ST6-IV [2B]	5	WA51	3 (0.8)	3 (100)
ST8-IV [2B]	8	WA5	3 (0.8)	0
ST953-IV [2B]	97	WA54	3 (0.8)	0
ST772-V [5C2]	1	Bengal Bay	2 (0.6)	2 (100)
ST1-V [5C2]	1		1 (0.3)	0
ST188-IV [2B]	1	WA38	1 (0.3)	0
ST5-IV [2B]	5	WA96	1 (0.3)	0
ST5-IV [2B]	5	WA71	1 (0.3)	1 (100)
ST5-V [5C2]	5	WA109	1 (0.3)	0
ST5-V [5C2]	5		1 (0.3)	0
ST835-IV [2B]	5	WA48	1 (0.3)	0
ST2471-V [5C2]	8	WA120	1 (0.3)	0
ST12-novel	12	WA59	1 (0.3)	0
ST30-V [5C2]	30	WA124	1 (0.3)	1 (100)
ST45-IV [2B]	45	WA75	1 (0.3)	0
ST59-IV [2B]	59	WA15	1 (0.3)	0
ST59-IV [2B]	59	WA55	1 (0.3)	1 (100)
ST72-IV [2B]	72	Korean Clone	1 (0.3)	0
ST577-IV [2B]	121	WA22	1 (0.3)	0
ST883-IV [2B]	Singleton	WA47	1 (0.3)	0
ST1303-IV [2B]	U	WA76	1 (0.3)	0
Total			355	223 (62.8)

PVL Panton Valentine leukocidin.

Percentage figures in parenthesis relate to CA-MRSA isolates.

ST93-IV [2B] (Qld CA-MRSA) accounted for 25.9% of MRSA ranging from 12.9% in Victoria to 47.8% in the Northern Territory (Table 9). PVL positive ST93-IV[2B] (Qld CA-MRSA) were typically resistant to the ß-lactam antimicrobials only (110/129) or additionally to erythromycin (17/129).

ST30-IV [2B] (SWP MRSA) accounted for 12.0% of MRSA ranging from 0% in the Australian Capital Territory to 25.7% in Queensland (Table 9). Overall 90% of PVL positive ST30-IV [2B] were resistant to the ß-lactam antimicrobials only.

ST1-IV [2B] (WA1) accounted for 9.6% of MRSA ranging from 0% in Tasmania and the Australian Capital Territory to 25.4% in Western Australia (Table 9). Typically PVL negative, 95.8% of isolates were non-multi-resistant (resistant to less than 3 ß-lactam antimicrobials).

The remaining 3 major CA-MRSA clones, ST45-V [5C2&5] (WA84), ST78-IV [2B] (WA2) and ST5-IV [2B] (WA3) accounted for 4.2%, 3.6% and 3.4% of MRSA respectively.

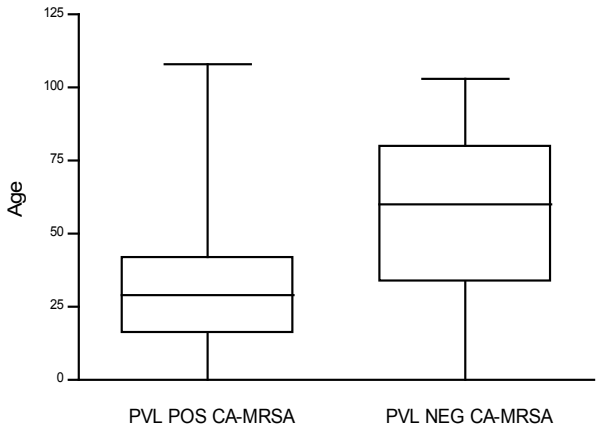
Overall, 94.4% of CA-MRSA were non-multi-resistant, with 61.4% of isolates resistant to ß-lactam antimicrobials only. However, 20 isolates (5.6% of CA-MRSA) were multi-resistant including 2 PVL positive ST772-V [5C2] (Bengal Bay MRSA) isolates, which, in addition to ß-lactam antimicrobials, were resistant to gentamicin, erythromycin, ciprofloxacin, and cotrimoxazole. Two CA-MRSA, ST188-IV [2B] (WA38) and ST8-IV [2B] (USA300), were resistant to 5 non-ß-lactam antimicrobials: gentamicin, erythromycin, ciprofloxacin, cotrimoxazole and tetracycline; and gentamicin, erythromycin, ciprofloxacin, mupirocin and tetracycline respectively.

PVL determinants were detected in 45.5% of MRSA:223 (62.8%) CA-MRSA (Table 8) and 4 ST22-IV [2B] (EMRSA-15) isolates. In addition to ST93-IV [2B] (Qld CA-MRSA) and ST30-IV [2B] (SWP MRSA), PVL-positive CA-MRSA clones included the international clones ST8-IV [2B] (USA300), ST59-V [5C2&5] (Taiwan MRSA) and ST772-V [5C2] (Bengal Bay MRSA). The mean age of patients (31.7 years: 95% CI 28.9–34.5) with a PVL positive CA-MRSA infection was significantly lower ($P<0.0001$) than the mean age of patients with a PVL negative CA-MRSA infection (55.7 years, 95% CI 50.7–60.6) (Figure 3).

Discussion

This survey demonstrates MRSA has become a significant burden in the Australian community. Over the 7 biennial AGAR community surveys (2000 to 2012), the percentage of *S. aureus* identified as MRSA has increased significantly ($P<0.0001$) by 6 percentage points over the 12-year period (11.5% in 2000 to 17.9% in 2012). Molecular typ-

Figure 3: Box plot of age of patients infected with Panton Valentine leukocidin positive and Panton Valentine leukocidin negative community-associated methicillin-resistant *Staphylococcus aureus*, Australia, 2012



Panton Valentine leukocidin positive community-associated methicillin-resistant *Staphylococcus aureus*: Mean age 31.7 years (95% CI: 28.9–34.5)

Panton Valentine leukocidin negative community-associated methicillin-resistant *Staphylococcus aureus*: Mean age 55.7 years (95% CI: 50.7–60.6)

Table 9: Proportion of methicillin-resistant *Staphylococcus aureus* characterised as community-associated methicillin-resistant *Staphylococcus aureus*, Australia, 2012, by state or territory

	ACT	NSW	NT	Qld	SA	Tas.	Vic.	WA	Aus.
ST93-IV [2B] (Qld)	25.0	21.5	47.8	37.6	33.3	22.2	12.9	23.8	25.9
ST30-IV [2B] (SWP)	0.0	8.7	13.0	25.7	9.5	22.2	7.1	6.3	12.0
ST1-IV [2B] (WA1)	0.0	6.4	4.3	8.9	14.3	0.0	5.9	25.4	9.6
ST45-V [5C2&5] (WA84)	0.0	2.9	0.0	2.0	2.4	0.0	15.3	0.0	4.2
ST78-IV [2B] (WA2)	25.0	0.6	0.0	1.0	7.1	11.1	2.4	14.3	3.6
ST5-IV [2B] (WA3)	25.0	1.7	0.0	5.0	2.4	0.0	4.6	4.8	3.4
Other	0.0	25.8	16.7	8.0	17.1	0.0	25.5	12.9	12.2

ing has shown this increase in community-onset MRSA has primarily been due to the emergence and expansion of non-multi-resistant clones.

In the 2012 study, resistance in MRSA to erythromycin, ciprofloxacin, tetracycline, gentamicin, clindamycin and cotrimoxazole significantly varied across regions. These differences can be explained by the different MRSA clones in circulation in each region; for example Aus-2/3 EMRSA (ST239-III), which is reliably resistant to gentamicin, erythromycin, tetracycline, ciprofloxacin and cotrimoxazole are commonly found in New South Wales, the Northern Territory and Victoria.

There were significant differences in the proportion of resistance to non-ß-lactam antimicrobials in MRSA associated with various patient types with gentamicin, tetracycline, ciprofloxacin, clindamycin, cotrimoxazole and fusidic acid resistance higher in hospital outpatients that other patient types. This is consistent with their having a higher proportion of healthcare-related acquisition.

In the 2012 study, apart from erythromycin, resistance to the non-ß-lactam antimicrobials amongst the MSSA was uncommon. Over the 7 AGAR surveys, no trends in resistance, increase or decrease, were evident for erythromycin, tetracycline, gentamicin or rifampicin. Nationally, small but significant increases were seen for clindamycin, ciprofloxacin, fusidic acid, high-level mupirocin and cotrimoxazole.

The mean age of patients with infections due to CA-MRSA strains (41 years; median 38 years) was found to be significantly lower ($P<0.0001$) than the mean age of patients with infections due to HA-MRSA strains (70 years; median 75 years). Although the percentage of *S. aureus* characterised as HA-MRSA in this survey (5.1%) was lower when compared with the 2010 survey (5.9%), ST22-IV [2B] (EMRSA-15) remains a major HA-MRSA clone in most Australian communities surveyed, accounting for 21.0% of all community-onset MRSA infections. Of continuing concern has been the rapid emergence of this clone in the community in Victoria (0% in 2002 to 21.2% in 2012), and New South Wales (18.0% in 2000 to 34.3% in 2012). In 2012, CA-MRSA accounted for 71.1% of MRSA and 12.5% of all *S. aureus*. Since 2000, the percentage of *S. aureus* characterised as CA-MRSA has more than doubled (5.3% in 2000). As in previous surveys although CA-MRSA was multi-clonal (32 clones,) 82.5% of strains could be characterised into 6 clones. ST93-IV [2B] (Qld CA-MRSA), a PVL-positive clone, remains the most frequently isolated CA-MRSA clone in the Australian community accounting for 36.3% of all CA-MRSA and 25.9% of all MRSA infections. Overall, 62.8% of CA-MRSA were PVL positive, a 21% increase when compared with the 2006 survey. The mean age of patients with PVL positive CA-MRSA infections (32 years; median 29 years) was significantly lower ($P<0.0001$) than the mean age of patients with PVL negative CA-MRSA infections (56 years; median 57 years). However, the increase in PVL-positive MRSA is not only due to the expansion of the ST93-IV [2B] clone but also due to ST30-IV [2B] (SWP MRSA) and due to the introduction of several international CA-MRSA clones including ST8-IV [2B] (USA300) ST59-V [5C2&5] (Taiwan CA-MRSA) and the hypervirulent multi-resistant ST772-V [5C2] (Bengal Bay). Four ST22-IV [2B] (EMRSA-15) isolates carrying the PVL determinant were also identified. Acquisition of the PVL determinant in this clone, which has been demonstrated to have enhanced transmission in the Australian community, continues to be a major public health concern.

In summary, resistance in MSSA remains uncommon with the exception of erythromycin and penicillin. Resistance in MRSA appears dynamic due to the success or decline of MRSA clones circulating in Australia. The national rate continues to rise in strains causing infections in people in the community. Since the initial AGAR community-onset *S. aureus* survey in 2000 there has been a significant increase in the percentage of patients with community onset MRSA infections in most regions of Australia, such that in 2012 one in 6 patients with a staphylococcal infection have MRSA and one in eight are infected with a CA-MRSA clone. This makes the empiric choice for the correct antibiotic therapy of community *S. aureus* infections increasingly difficult. Of further concern is that this increase in MRSA has primarily been due to the expansion of the PVL positive clones such as ST93-IV [2B] (Qld CA-MRSA) and ST30-IV [2B] (SWP CA-MRSA). This shift in the molecular epidemiology of MRSA clones in the Australian community will potentially increase the number of SSTIs in young Australians. As SSTIs caused by PVL-positive *S. aureus* frequently results in hospitalisation the emergence of these strains in the community as well as the detection of PVL-positive HA-MRSA (EMRSA-15) is a major health concern.

A full detailed report on this study may be found on the AGAR web site: (http://www.antimicrobial-resistance.com/) under AMR surveillance.

Acknowledgements

This study was primarily funded by a grant from the Australian Government Department of Health.

We gratefully acknowledge Tam Le from the Australian Collaborating Centre for Enterococcus and Staphylococcus Species (ACCESS) Typing and Research, School of Biomedical Sciences, Curtin University; and the Western Australia Genome Resource Centre, Department of Clinical Immunology and Biochemical Genetics, Royal Perth Hospital for the molecular typing of MRSA.

Agar participants

The members of AGAR for 2012 were:

Australian Capital Territory

Peter Collignon and Susan Bradbury, The Canberra Hospital

New South Wales

Tom Gottlieb and Graham Robertson, Concord Hospital

Miriam Paul and Richard Jones, Douglass Hanly Moir Pathology

James Branley and Donna Barbaro, Nepean Hospital

George Kotsiou and Peter Huntington, Royal North Shore Hospital

Sebastian van Hal and Bradley Watson, Royal Prince Alfred Hospital

Iain Gosbell and Annabelle LeCordier, South West Area Pathology Service

David Mitchell and Lee Thomas, Westmead Hospital

Northern Territory

Jann Hennessy and Rob Baird, Royal Darwin Hospital

Queensland

Enzo Binotto and Bronwyn Thomsett, Pathology Queensland Cairns Base Hospital

Graeme Nimmo and Narelle George, Pathology Queensland Central Laboratory

Petra Derrington and Sharon Dal-Cin, Pathology Queensland Gold Coast Hospital

Chris Coulter and Tobin Hillier, Pathology Queensland Prince Charles Hospital

Naomi Runnegar and Joel Douglas, Pathology Queensland Princess Alexandra Hospital

Jenny Robson and Georgia Peachey, Sullivan Nicolaides Pathology

South Australia

Kelly Papanoum and Nicholas Wells, SA Pathology, Flinders Medical Centre

Morgyn Warner and Fleur Manno, SA Pathology, Institute of Medical and Veterinary Science

John Turnidge and Jan Bell, SA Pathology, Women's and Children's Hospital

Tasmania

Kathy Wilcox, Launceston General Hospital

Louise Cooley and Rob Peterson, Royal Hobart Hospital

Victoria

Denis Spelman and Michael Huysmans, Alfred Hospital

Benjamin Howden and Peter Ward, Austin Hospital

Tony Korman and Despina Kotsanas, Monash Hospital Medical Centre

Sue Garland and Gena Gonis, Royal Women's Hospital

Mary Jo Waters and Linda Joyce, St Vincent's Hospital

Western Australia

David McGechie and Rebecca Wake, PathWest Laboratory Medicine, WA Fremantle Hospital

Barbara Henderson and Ronan Murray, PathWest Laboratory Medicine, WA Queen Elizabeth II Hospital

Owen Robinson and Geoffrey Coombs, PathWest Laboratory Medicine, WA Royal Perth Hospital

Sudha Pottumarthy-Boddu and Fay Kappler, St John of God Pathology

Author details

Dr Geoffrey W Coombs[1,2]
Ms Denise A Daley[3]
Ms Julie C Pearson[1,2]
Prof Graeme R Nimmo[4,5]
Prof Peter J Collignon[6]
Prof Mary-Louise McLaws[7]
Dr James O Robinson[1,2]
Prof John D Turnidge[8,9]

1. Australian Collaborating Centre for Enterococcus and Staphylococcus Species (ACCESS) Typing and Research, School of Biomedical Sciences, Curtin University, Perth, Western Australia
2. Department of Microbiology and Infectious Diseases, PathWest Laboratory Medicine-WA, Royal Perth Hospital, Perth, Western Australia
3. Australian Group on Antimicrobial Resistance, Royal Perth Hospital, Perth, Western Australia
4. Division of Microbiology, Pathology Queensland Central Laboratory, Herston Hospitals Campus, Herston, Queensland
5. School of Medicine, Griffith University, Gold Coast, Queensland
6. Infectious Diseases Unit and Microbiology Department, The Canberra Hospital, Garran, Australian Capital Territory
7. Healthcare Associated Infection and Infectious Diseases Control, SPHCM, UNSW Australia, Sydney, New South Wales
8. SA Pathology, Department of Microbiology and Infectious Diseases, Women's and Children's Hospital, North Adelaide, South Australia
9. Departments of Pathology, Paediatrics and Molecular and Biomedical Sciences, University of Adelaide, Adelaide, South Australia

Corresponding author: Dr Geoffrey Coombs, Australian Collaborating Centre for Enterococcus and Staphylococcus Species (ACCESS) Typing and Research, School of Biomedical Sciences, Curtin University, Perth, Western Australia, 8000. Telephone: +61 8 9224 2446. Facsimile: +61 8 9224 1989. Email: Geoff.Coombs@curtin.edu.au

References

1. Turnidge JD, Kotsanas D, Munckhof W, Roberts S, Bennett CM, Nimmo GR, et al. *Staphylococcus aureus* bacteraemia: a major cause of mortality in Australia and New Zealand. *Med J Aust* 2009;191(7):368–373.
2. Collignon P, Nimmo GR, Gottlieb T, Gosbell IB, Australian Group on Antimicrobial Resistance. *Staphylococcus aureus* bacteremia, Australia. *Emerg Infect Dis* 2005;11(4):554–561.
3. Rountree PM, Beard MA. Hospital strains of *Staphylococcus aureus,* with particular reference to methicillin-resistant strains. *Med J Aust* 1968;2(26):1163–1168.
4. Pavillard R, Harvey K, Douglas D, Hewstone A, Andrew J, Collopy B, et al. Epidemic of hospital-acquired infection due to methicillin-resistant *Staphylococcus aureus* in major Victorian hospitals. *Med J Aust* 1982;1(11):451–454.
5. Rountree PM. History of staphylococcal infection in Australia. *Med J Aust* 1978;2(12):543–546.
6. Riley TV, Pearman JW, Rouse IL. Changing epidemiology of methicillin-resistant *Staphylococcus aureus* in Western Australia. *Med J Aust* 1995;163(8):412–414.
7. Udo EE, Pearman JW, Grubb WB. Genetic analysis of community isolates of methicillin-resistant *Staphylococcus aureus* in Western Australia. *J Hosp Infect* 1993;25(2):97–108.
8. Maguire GP, Arthur AD, Boustead PJ, Dwyer B, Currie BJ. Emerging epidemic of community-acquired methicillin-resistant *Staphylococcus aureus* infection in the Northern Territory. *Med J Aust* 1996;164(12):721–723.
9. Collignon P, Gosbell I, Vickery A, Nimmo G, Stylianopoulos T, Gottlieb T. Community-acquired methicillin-resistant *Staphylococcus aureus* in Australia. Australian Group on Antimicrobial Resistance. *Lancet* 1998;352(9122):145–146.
10. Gosbell IB, Mercer JL, Neville SA, Crone SA, Chant KG, Jalaludin BB, et al. Non-multiresistant and multiresistant methicillin-resistant *Staphylococcus aureus* in community-acquired infections. *Med J Aust* 2001;174(12):627–630.
11. Nimmo GR, Schooneveldt J, O'Kane G, McCall B, Vickery A. Community acquisition of gentamicin-sensitive methicillin-resistant *Staphylococcus aureus* in southeast Queensland, Australia. J *Clin Microbiol* 2000;38(11):3926–3931.
12. Gosbell IB, Mercer JL, Neville SA, Chant KG, Munro R. Community-acquired, non-multiresistant oxacillin-resistant *Staphylococcus aureus* (NORSA) in South Western Sydney. *Pathology* 2001;33(2):206–210.
13. Munckhof WJ, Schooneveldt J, Coombs GW, Hoare J, Nimmo GR. Emergence of community-acquired methicillin-resistant *Staphylococcus aureus* (MRSA) infection in Queensland, Australia. *Int J Infect Dis* 2003;7(4):259–264.
14. Coombs GW, Nimmo GR, Pearson JC, Christiansen KJ, Bell JM, Collignon PJ, et al. Prevalence of MRSA strains among *Staphylococcus aureus* isolated from outpatients, 2006. *Commun Dis Intell* 2009;33(1):10–20.
15. Coombs GW, Monecke S, Pearson JC, Tan HL, Chew YK, Wilson L, et al. Evolution and diversity of community-associated methicillin-resistant *Staphylococcus aureus* in a geographical region. *BMC Microbiol* 2011;11:215.
16. Nimmo GR, Bell JM, Collignon PJ. Fifteen years of surveillance by the Australian Group for Antimicrobial Resistance (AGAR). *Commun Dis Intell* 2003;27(Suppl):S47–S54.
17. Nimmo GR, Bell JM, Mitchell D, Gosbell IB, Pearman JW, Turnidge JD. Antimicrobial resistance in *Staphylococcus aureus* in Australian teaching hospitals, 1989–1999. *Microb Drug Resist* 2003;9(2):155–160.
18. Turnidge J, Lawson P, Munro R, Benn R. A national survey of antimicrobial resistance in *Staphylococcus aureus* in Australian teaching hospitals. *Med J Aust* 1989;150(2):65, 69–72.
19. Turnidge JD, Nimmo GR, Francis G. Evolution of resistance in *Staphylococcus aureus* in Australian teaching hospitals. Australian Group on Antimicrobial Resistance (AGAR). *Med J Aust* 1996;164(2):68–71.
20. Nimmo GR, Coombs GW, Pearson PC, O'Brien FG, Christiansen KJ, Turnidge JD, et al. MRSA in the Australian community: an evolving epidemic. Med *J Aust* 2006;184(8):384–388.
21. Coombs GW, Nimmo GR, Bell JM, Huygens F, O'Brien FG, Malkowski MJ, et al. Genetic diversity among community methicillin-resistant *Staphylococcus aureus* strains causing outpatient infections in Australia. *J Clin Microbiol* 2004;42(10):4735–4743.

22. Coombs GW, Nimmo GR, Pearson JC, Christiansen KJ, Bell JM, Collignon PJ, et al. Prevalence of MRSA strains among *Staphylococcus aureus* isolated from outpatients, 2006. *Commun Dis Intell* 2009;33(1):10–20.

23. Clinical and Laboratory Standards Institute. *Performance standards for antimicrobial susceptibility testing*. Twenty-second informational supplement M100-S22. Villanova, PA, USA 2012.

24. O'Brien FG, Udo EE, Grubb WB. Contour-clamped homogeneous electric field electrophoresis of *Staphylococcus aureus*. *Nat Protoc* 2006;1(6):3028–3033.

25. 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(3):1008–1015.

26. Goh SH, Byrne SK, Zhang JL, Chow AW. Molecular typing of *Staphylococcus aureus* on the basis of coagulase gene polymorphisms. *J Clinical Microbiol* 1992;30(7):1642–1645.

27. Coombs GW, Monecke S, Ehricht R, Slickers P, Pearson JC, Tan HL, et al. Differentiation of clonal complex 59 community-associated methicillin-resistant *Staphylococcus aureus* in Western Australia. *Antimicrob Agent Chemother* 2010;54(5):1914–1921.

28. Fey PD, Said-Salim B, Rupp ME, Hinrichs SH, Boxrud DJ, Davis CC, et al. Comparative molecular analysis of community– or hospital-acquired methicillin-resistant *Staphylococcus aureus*. *Antimicrob Agent Chemother* 2003;47(1):196–203.

29. Enright MC, Robinson DA, Randle G, Feil EJ, Grundmann H, Spratt BG. The evolutionary history of methicillin-resistant *Staphylococcus aureus* (MRSA). *Proc Natl Acad Sci U S A* 2002;99(11):7687–7692.

30. Classification of staphylococcal cassette chromosome mec (SCC*mec*): guidelines for reporting novel SCC*mec* elements. *Antimicrob Agent Chemother*. 2009;53(12):4961–4967.