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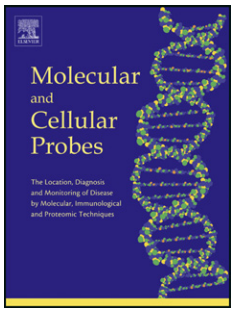

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Distribution of SCC*mec*-associated phenol-soluble modulin in staphylococci

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Abstract

The recently described phenol-soluble modulin *PSM-mec* was detected in *S. aureus, S. epidermidis, S. fleuretti, S. hominis, S. pseudintermedius, S. saprophyticus, S. simulans* and *S. vitulinus* from different hosts (humans, goats, dogs, cats, pigs, cattle and turkeys). It was identified in isolates harbouring SCC*mec* types II, IIA, IIB, IID, III, VIII and in some irregular or truncated elements.

Staphylococcus aureus expresses, among other virulence factors, phenol-soluble modulins (PSMs). PSMs are small, amphipathic and α-helical peptides that play a role in immune evasion as pro-inflammatory cytolytic toxins [1]. Recently, Queck *et al*. described a hitherto unknown PSM gene [2] in methicillin-resistant *S. aureus* (MRSA). This gene, *PSM-mec*, partially overlaps a putative virulence-modulating open reading frame, "*fudoh*" [3]. In contrast to previously known core-genomic PSM genes, *PSM-mec* is localised within SCC*mec* elements [4, 5] which comprise the gene responsible for methicillin resistance, *mecA*, recombinase genes, regulatory elements and, variably, additional resistance genes [5-8]. *PSM-mec* is the only known virulence factor associated with these elements. It has been found in *S. aureus* SCC*mec* types II, III and VIII and *S. epidermidis* SCC*mec* types II and III [2] as well as in *S. cohnii* (see GenBank GU370073.2) *S. saprophyticus* (AB353724.1), *S. sciurii* (Y13095.1, Y13096.1, AB547234.1, AB547235.1, AB547236.1) and *S. vitulinus* (AB546780.1).

The aim of this study was to screen staphylococcal isolates for the presence of *PSM-mec* in order to obtain more data on its distribution. 102 *S. aureus* isolates and 38 coagulase-negative staphylococci were genotyped using DNA microarrays (StaphyType, Alere Technologies GmbH, Jena, Germany, [7, 9]). This allowed characterising their SCC*mec* elements [10]. In addition, SCC*mec* elements of some isolates have previously been studied comprehensively [11, 12].

PCR primers pr_psmMEC_02 (5'-CGAAAGCCTGAATGCAAGTCT-3') and pr_psmMEC_03 (5'-GGATTTCACTGGTGTTATTACAAGC-3') were used to detect *PSM-mec*. Reaction conditions included an initial denaturation (2 min at 96°C) followed by 35 cycles (20 sec at 96°C, 20 sec at 70°C and 20 sec at 72°C). A second PCR covered a region spanning from *xylR* to *PSM-mec* (pr_psmMEC_02 and pr_xylR_04, 5'-AAGCGTCATCTTCTCATTTAGTTGA-3') and a third PCR covered the region from *PSM-mec* to *mecR1* (pr_mecR_01, 5'-CCAGAAAGTAAACAACGATATTCACC-3' and pr_psmMEC_03). Both reactions comprised denaturation (2 min at 96°C) followed by 35 cycles (20 sec at 96°C, 20 sec at 55°C and 70 sec at

72°C). *S. aureus* Mu50 (ST5-MRSA-II, GenBank BA000017), N315 (ST5-MRSA-II, BA000018) and Sanger MRSA 252 (ST36-MRSA-II, BX571856) were used as positive as well as NCTC 8325 (ST8-MSSA, CP000253,), USA300-FPR3757 (ST8-MRSA-IV, CP000255), Sanger MSSA 476 (ST1-MSSA, BX571857), COL (ST250-MRSA-I, CP000046,) and MW2 (ST1-MRSA-IV, BA00003) as negative controls.

The results are summarised in Table 1. *PSM-mec* (as detected using the primers pr_psmMEC_02/pr_psmMEC_03) was detected in association with *mecI* and/or *xylR, i.e.*, in isolates harbouring SCC*mec* types II, IIA, IIB, IID, III, VIII or irregular/composite elements related to SCC*mec* types IIA/B and III. *PSM-mec* was not restricted to *S. aureus*. It was detected in *S. epidermidis, S. fleuretti, S. hominis, S. pseudintermedius, S. saprophyticus, S. simulans* and *S. vitulinus.* The presence of *PSM-mec* did not depend on the host species, and was found in isolates from humans, dogs, cats, goats, cattle, pigs and turkeys. *PSM-mec* was absent in SCC*mec* types I, IIC, IIE, IV, V or XI and in SCC elements lacking the *mec* complex. Some isolates, including CC12-MRSA, WA-MRSA-59, harboured SCC*mec* elements comprising *mecR1* but lacking *xylR*. These strains were *PSM-mec*-negative.

There was no evidence for alternative locations of *PSM-mec*. Isolates lacking SCC*mec* elements, or harbouring SCC*mec* types I, IIC, IIE, IV and V were *PSM-mec*-negative regardless of clonal complex or species affiliation. A PCR covering the region from *xylR* to *PSM-mec* using primers pr_psmMEC_02/pr_xylR_04 yielded results which were in all cases but one in accordance with the *PSM-mec* PCR. In a *PSM-mec*-positive CC5-MRSA, *xylR* was absent, and primer pair pr_psmMEC_02/pr_xylR_04 did not yield a result.

Results of a third PCR (pr_psmMEC/pr_mecR_01) differed from those of the other two PCRs by yielding negative results for ST8-MRSA-IIA or -IID, and for deletion variants of this strain. This can be attributed to the insertion of a 1865 bp long IS1182 element and between the primers,

within *mecI* [11, 12]. It was also negative in a *S. vitulinus* strain, which was *mecR1*-negative by array hybridisation.

Associations of virulence factors with mobile genetic elements (MGEs) conferring drug resistance have been observed in *E. coli* [13, 14] and enterococci [15], but in staphylococci, the connection of PSM-*mec* to SCC*mec* is rather unique [2]. *PSM-mec* can be found in diverse staphylococcal species, and these staphylococci originated from different host species. This suggests that the effects of *PSM-mec* are rather independent from genomic background and that they are not restricted to specific hosts, although further functional studies on that issue are required.

A variety of SCC*mec* elements have been identified [4]. Similar to other MGEs, they undergo evolutionary changes independent of their hosts [16] and may compete against other elements with similar properties and integration sites. Resistance-associated MGEs provide an advantage only in the presence of antibiotics. Otherwise, they can even be disadvantageous [17, 18]. Thus, for such an element it is necessary to ensure its maintenance in the absence of selective pressure, *e.g.,* if an antibiotic therapy was discontinued. It can evolve to cause no or only minimal disadvantage in the absence of antibiotics, as it is the case for SCC*mec* type IV [17, 18], to impose a selective pressure by itself [19] or, alternatively, to encode a factor conferring a selective advantage independently of the presence of antibiotics. Since experimental data show an influence of PSM-*mec* on virulence and/or biofilm formation [2, 20], its presence might be a key factor for the evolutionary success (*i.e.*, the pandemic spread and abundance) of SCC*mec* II and III elements and their host strains such as ST239-MRSA-III and ST5/ST225-MRSA-II.

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TABLES AND FIGURES

Table 1: Details of tested strains.

Figure 1: Overview over approximate gene and primer localisations, based on positions within the N315 genomes.

REFERENCES

1. Wang R, Braughton KR, Kretschmer D, Bach T-HL, Queck SY, Li M, et al. Identification of novel cytolytic peptides as key virulence determinants for community-associated MRSA. Nat Med 2007;13:1510-1514.
2. Queck SY, Khan BA, Wang R, Bach TH, Kretschmer D, Chen L, et al. Mobile genetic element-encoded cytolysin connects virulence to methicillin resistance in MRSA. PLoS Pathog 2009;5:e1000533.
3. Kaito C, Omae Y, Matsumoto Y, Nagata M, Yamaguchi H, Aoto T, et al. A novel gene, fudoh, in the SCC*mec* region suppresses the colony spreading ability and virulence of *Staphylococcus aureus*. PLoS One 2008;3:e3921.
4. IWG-SCC. Classification of staphylococcal cassette chromosome *mec* (SCC*mec*): guidelines for reporting novel SCC*mec* elements. Antimicrob Agents Chemother 2009;53:4961-7.
5. Ito T, Katayama Y, Asada K, Mori N, Tsutsumimoto K, Tiensasitorn C, et al. Structural comparison of three types of staphylococcal cassette chromosome *mec* integrated in the chromosome in methicillin-resistant *Staphylococcus aureus*. Antimicrob Agents Chemother 2001;45:1323-36.
6. Oliveira DC, Wu SW, de Lencastre H. Genetic organization of the downstream region of the *mecA* element in methicillin-resistant *Staphylococcus aureus* isolates carrying different polymorphisms of this region. Antimicrob Agents Chemother 2000;44:1906-10.
7. Monecke S, Coombs G, Shore AC, Coleman DC, Akpaka P, Borg M, et al. A Field Guide to Pandemic, Epidemic and Sporadic Clones of Methicillin-Resistant *Staphylococcus aureus*. PLoS ONE 2011;6:e17936.
8. Shore AC, Deasy EC, Slickers P, Brennan G, O'Connell B, Monecke S, et al. Detection of Staphylococcal Cassette chromosome *mec* type XI encoding highly divergent *mecA*, *mec*I, *mec*R1, *blaZ* and *ccr* genes in human clinical clonal complex 130 methicillin-resistant *Staphylococcus aureus*. Antimicrob Agents Chemother 2011;55.
9. Monecke S, Slickers P, Ehricht R. Assignment of *Staphylococcus aureus* isolates to clonal complexes based on microarray analysis and pattern recognition. FEMS Immunol Med Microbiol 2008;53:237–251.
10. Coombs GW, Monecke S, Ehricht R, Slickers P, Pearson JC, Tan HL, et al. Differentiation of CC59 community-associated methicillin-resistant *Staphylococcus aureus* in Western Australia. Antimicrob Agents Chemother 2010;54:1914-21.
11. Shore AC, Rossney AS, O'Connell B, Herra CM, Sullivan DJ, Humphreys H, et al. Detection of staphylococcal cassette chromosome *mec*-associated DNA segments in multiresistant methicillin-susceptible *Staphylococcus aureus* (MSSA) and identification of Staphylococcus epidermidis ccrAB4 in both methicillin-resistant S. aureus and MSSA. Antimicrob Agents Chemother 2008;52:4407-19.
12. Shore A, Rossney AS, Keane CT, Enright MC, Coleman DC. Seven novel variants of the staphylococcal chromosomal cassette *mec* in methicillin-resistant *Staphylococcus aureus* isolates from Ireland. Antimicrob Agents Chemother 2005;49:2070-83.
13. Johnson TJ, Giddings CW, Horne SM, Gibbs PS, Wooley RE, Skyberg J, et al. Location of increased serum survival gene and selected virulence traits on a conjugative R plasmid in an avian *Escherichia coli* isolate. Avian Dis 2002;46:342-52.
14. Boerlin P, Travis R, Gyles CL, Reid-Smith R, Janecko N, Lim H, et al. Antimicrobial resistance and virulence genes of *Escherichia coli* isolates from swine in Ontario. Appl Environ Microbiol 2005;71:6753-61.
15. Clewell DB, Victoria Francia M, Flannagan SE, An FY. Enterococcal plasmid transfer: sex pheromones, transfer origins, relaxases, and the *Staphylococcus aureus* issue. Plasmid 2002;48:193-201.
16. Novick RP. Mobile genetic elements and bacterial toxinoses: the superantigen-encoding pathogenicity islands of *Staphylococcus aureus*. Plasmid 2003;49:93-105.
17. Lee SM, Ender M, Adhikari R, Smith JMB, Berger-Bachi B, Cook GM. Fitness cost of staphylococcal cassette chromosome *mec* in methicillin-resistant *Staphylococcus aureus* by way of continuous culture. Antimicrob Agents Chemother 2007;51:1497-1499.
18. Ender M, McCallum N, Adhikari R, Berger-Bachi B. Fitness Cost of SCC*mec* and Methicillin Resistance Levels in *Staphylococcus aureus*. Antimicrob Agents Chemother 2004;48:2295-2297.
19. Ellington MJ, Woodford N. Fluoroquinolone resistance and plasmid addiction systems: self-imposed selection pressure? J Antimicrob Chemother 2006;57:1026-9.
20. Kaito C, Saito Y, Nagano G, Ikuo M, Omae Y, Hanada Y, et al. Transcription and Translation Products of the Cytolysin Gene psm-*mec* on the Mobile Genetic Element SCC*mec* Regulate *Staphylococcus aureus* Virulence. PLoS Pathog 2011;7:e1001267 EP -.
21. Fessler AT, Billerbeck C, Kadlec K, Schwarz S. Identification and characterization of methicillin-resistant coagulase-negative staphylococci from bovine mastitis. J Antimicrob Chemother 2010;65:1576-82.
22. Descloux S, Rossano A, Perreten V. Characterization of new staphylococcal cassette chromosome *mec* (SCC*mec*) and topoisomerase genes in fluoroquinolone- and methicillin-resistant *Staphylococcus pseudintermedius*. J Clin Microbiol 2008;46:1818-23.
23. Schwarz S, Kadlec K, Strommenger B. Methicillin-resistant *Staphylococcus aureus* and *Staphylococcus pseudintermedius* detected in the BfT-GermVet monitoring programme 2004-2006 in Germany. J Antimicrob Chemother 2008;61:282-5.
24. Schwarz S, Alesik E, Werckenthin C, Grobbel M, Lübke-Becker A, Wieler L, et al. Antimicrobial susceptibility of coagulase-positive and coagulase-variable Staphylococci from various indications of swine, dogs and cats as determined in the BfT-GermVet monitoring program 2004-2006. Berl Munch Tierarztl Wochenschr 2007;120:372-9.
25. Nienhoff U, Kadlec K, Chaberny IF, Verspohl J, Gerlach GF, Schwarz S, et al. Methicillin-resistant *Staphylococcus pseudintermedius* among cats admitted to a veterinary teaching hospital. Vet Microbiol 2011;153:414-6.
26. Nienhoff U, Kadlec K, Chaberny IF, Verspohl J, Gerlach GF, Kreienbrock L, et al. Methicillin-resistant *Staphylococcus pseudintermedius* among dogs admitted to a small animal hospital. Vet Microbiol 2011;150:191-7.
27. Scicluna E, Shore A, Thuermer A, Ehricht R, Slickers P, Borg M, et al. Characterisation of MRSA from Malta and the description of a Maltese epidemic MRSA strain. Eur J Clin Microbiol Infect Dis 2010;29:163-170.
28. Walther B, Monecke S, Ruscher C, Friedrich AW, Ehricht R, Slickers P, et al. Comparative molecular analysis substantiates a zoonotic potential of equine Methicillin-resistant *Staphylococcus aureus* (MRSA). J Clin Microbiol 2009;47:704–710.

Description of the SCC*mec* element				Species	Strain designations	Host species	No. of isolates tested	*PSM-mec PCR*	*PSM-mec /xylR PCR*	*mecR1 / PSM-mec PCR*
SCC*mec* type	***mec*-complex**	**Recombinase genes**	**Accessory genes (variable genes in brackets)**							
None	-	-	-	*S. aureus*	CC8-MSSA, including NCTC 8325	Human	2	-	-	-
				S. aureus	CC30-MSSA [PVL+], ATCC25923	Human	1	-	-	-
				S. aureus	CC398-MSSA	Turkey	1	-	-	-
	-	*ccrA/B-4*	ACME	*S. aureus*	CC8-MSSA	Human	1	-	-	-
	-	*ccrA/B-2*	*kdp*-SCC-locus	*S. aureus*	CC182-MSSA	Human	2	-	-	-
	-	*ccrA/B-1*	Q6GD50=fusidic acid resistance	*S. aureus*	CC1-MSSA-SCC*fus*, Sanger MSSA476	Human	1	-	-	-
I (1B)	*mecA, ugpQ, Delta mecR1*	*ccrA/B-1*	(*pls*-SCC)	*S. aureus*	CC8/ST250-MRSA-I, Early/Ancestral MRSA, including COL	Human	3	-	-	-
				S. aureus	CC8/ST247-MRSA-I, North German/Iberian EMRSA	Human	3	-	-	-
I (1B) variant	*mecA, ugpQ, Delta mecR1*	*(ccrA-1), ccrB-1*	(*pls*-SCC)	*S. aureus*	CC5-MRSA-I var., WA-MRSA-18/21/48	Human	3	-	-	-
II (2A)	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-2*	*kdp*-SCC-locus	*S. aureus*	ST5/ST225-MRSA-II, Rhine-Hesse/UK-EMRSA-3, including Mu50 and N315	Human	12	+	+	+
				S. aureus	CC30/ST36-MRSA-II, UK-EMRSA-16, including Sanger MRSA252 and ATCC43300	Human	3	+	+	+
				S. aureus	CC45-MRSA-II, USA600 (NARSA22)	Human	1	+	+	+
II/composite	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-2, ccrB-4*	*kdp*-SCC-locus	*S. epidermidis*	NARSA101, ATCC 35984	Human	1	+	+	+
IIA	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-2, ccrA/B-4*	*mer*-locus, *aadD*	*S. aureus*	ST8-MRSA-IIA, Irish AR13/14	Human	1	+	+	-
IIB	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-2, ccrA/B-4*	*mer*-locus	*S. aureus*	ST8-MRSA-IIB, Irish AR13/14	Human	1	+	+	+
IIC	*mecA, ugpQ, mecR1*	*ccrA/B-2, ccrA/B-4*	*mer*-locus, *aadD*	*S. aureus*	ST8-MRSA-IIC, Irish AR13/14	Human	2	-	-	-
IID	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-2, ccrA/B-4*	(*mer*-locus)	*S. aureus*	ST8-MRSA-IID, Irish AR13/14	Human	4	+	+	-
IIE	*mecA, ugpQ, mecR1*	*ccrA/B-2, ccrA/B-4*	*mer*-locus	*S. aureus*	ST8-MRSA-IIE, Irish AR13/14	Human	1	-	-	-

Description of the SCC*mec* element				**Species**	**Strain designations**	**Host species**	**No. of isolates tested**	***PSM-mec PCR***	***PSM-mec /xylR PCR***	***mecR1 / PSM-mec PCR***
III (3A)	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-3,(ccrC)*	(*mer*-locus, *tetK*)	*S. aureus*	CC5-MRSA-III	Turkey	3	+	+	+
				S. aureus	CC8/ST239-MRSA-III, Vienna/Hungarian/Brazilian Clone	Human	17	+	+	+
				S. sapro-phyticus	(from [21])	Cattle	1	+	+	+
				S. hominis	-	Human	1	+	+	+
III/composite	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-3, ccrA/B-4*	-	*S. epidermidis*	-	Human	3	+	+	+
				S. simulans		Human	1	+	+	+
"II-III" [22]	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-3*		*S. pseudo-intermedius*	KM1381 (from [22])	Dog	1	+	+	+
III or "II-III" [22]	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-3*		*S. pseudo-intermedius*	(from [23-26])	Cats, dogs	20	+	**+ (in 19/20)**	+
IV (2B)	*mecA, ugpQ, Delta mecR1*	*ccrA/B-2*		*S. aureus*	CC1-MRSA-IV [PVL+], MW2/USA400	Human	1	-	-	-
				S. aureus	CC8-MRSA-IV, UK-EMRSA-14/WA-MRSA-5	Human	4	-	-	-
				S. aureus	CC8-MRSA-IV, Lyon Clone/UK-EMRSA-2	Human	1	-	-	-
				S. aureus	CC8-MRSA-IV, UK-EMRSA-6	Human	1	-	-	-
				S. aureus	ST22-MRSA-IV, Barnim/EMRSA-15	Human	1	-	-	-
				S. aureus	ST30-MRSA-IV [PVL+], Southwest Pacific Clone	Human	1	-	-	-
				S. aureus	ST30-MRSA-IV	Human	1	-	-	-
				S. aureus	ST59-MRSA-IV [PVL+], WA MRSA-55/56	Human	1	-	-	-
				S. aureus	ST80-MRSA-IV, European Clone	Human	1	-	-	-
				S. aureus	ST93-MRSA-IV [PVL+], Queensland Clone	Human	1	-	-	-
				S. epidermidis	-	Human	1	-	-	-

Description of the SCC*mec* element				**Species**	**Strain designations**	**Host species**	**No. of isolates tested**	***PSM-mec PCR***	***PSM-mec /xylR PCR***	***mecR1 / PSM-mec PCR***
IV/composite	*mecA, ugpQ, Delta mecR1*	*ccrA/B-2*	ACME	*S. aureus*	ST8-MRSA-IV, USA300, FPR3757	Human	1	–	–	–
IV/composite	*mecA, ugpQ, Delta mecR1*	*ccrA/B-2*, *ccrA/B*-GU066221	Q6GD50	*S. aureus*	CC5-MRSA-IVvar, "Maltese Clone" [27]	Human	1	–	–	–
IV/composite	*mecA, ugpQ, Delta mecR1*	*(ccrA/B-2)*, *ccrA/B-4*	-	*S. aureus*	ST8-MRSA-(IV+*ccrA/B*-4), UK-EMRSA-12/13, Irish AR43	Human	3	–	–	–
V_T (5C2&5)	*mecA, ugpQ*	*ccrC*	-	*S. aureus*	ST59/ST952-MRSA-V_T [PVL+], Taiwan Clone	Human	2	–	–	–
				S. aureus	ST398-MRSA-V, from [28]	Horse	1	–	–	–
VIII (4A)	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-4*	-	*S. aureus*	CC8-MRSA-VIII	Human	1	+	+	+
				S. aureus	CC361-MRSA-VIII, WA-MRSA-28	Human	1	+	+	+
XI	Divergent *mecA, mecI, mecR1* [8]	Divergent *ccrA1/B3*	Arsenic resistance operon	*S. aureus*	CC130-MRSA-XI, from [8]	Human	2	–	–	–
Irregular SCC*mec* elements	*mecA, ugpQ*	*ccrC, ccrA/B-4*	-	*S. aureus*	ST779-MRSA	Human	1	–	–	–
	mecA, ugpQ, Delta mecR1	*ccrA/B-2, ccrC*	-	*S. aureus*	CC8/ST254-MRSA-IV/V, UK-EMRSA-10/Hannover EMRSA	Human	1	–	–	–
	mecA, ugpQ, Delta mecR1	-	*mer*-locus	*S. aureus*	CC8/ST254-MRSA, Hannover EMRSA	Human	1	–	–	–
	mecA, ugpQ, mecR1	*ccrC*	-	*S. aureus*	CC5-MRSA	Pig	2	–	–	–
	mecA, ugpQ, mecR1, mecI	*ccrA/B-2, ccrA/B-4*	*mer*-locus	*S. epidermidis*	-	Human	1	–	–	–
	mecA, ugpQ, mecR1, mecI	-	-	*S. aureus*	CC12-MRSA, WA-MRSA-59	Human	1	–	–	–
	mecA, ugpQ, mecR1, mecI	-	-	*S. aureus*	CC5-MRSA	Pig	5	+	–	+
	mecA, ugpQ, mecR1, mecI	-	-	*S. sapro-phyticus*	-	Human	1	–	–	–
	mecA, ugpQ, mecR1, mecI	*ccrA/B-1*	-	*S. hominis*	-	Human	1	–	–	–
	mecA, xylR	*ccrA/B-3*		*S. vitulinus*	(from [21])	Cattle	1	+	+	–
	mecA, ugpQ, mecR1, mecI	*ccrA/B-4*	-	*S. hominis*	-	Human	1	–	–	–

Description of the SCC*mec* element				**Species**	**Strain designations**	**Host species**	**No. of isolates tested**	***PSM-mec PCR***	***PSM-mec /xylR PCR***	***mecR1 / PSM-mec PCR***
Irregular or truncated II/ III	*mecA, ugpQ, mecR1, mecI, xylR*	-	-	*S. aureus*	CC398-MRSA	Turkey	1	+	+	+
				S. aureus	CC9-MRSA	Pig	1	+	+	+
			-	*S. fleuretti*	(including ATCC BAA-274)	Goat milk, humans with goat contact	3	+	+	+
	mecA, ugpQ, mecR1, mecI, xylR	*ccrC*	-	*S. sapro-phyticus*	-	Human	1	+	+	+
Truncated II A/B/D or VIII	*mecA, ugpQ, mecR1, mecI, xylR*	*ccrA/B-4*	*mer*-locus	*S. aureus*	CC8-MRSA-VIII or ST8-MRSA-II A, Irish AR13/14, *ccrA/B-2* deletion variant	Human	1	+	+	-
Truncated II D	*mecI, xylR*	*ccrA/B-2, ccrA/B-4*	*mer*-locus	*S. aureus*	ST8-MSSA, *mecA*-neg. deletion variant of ST8-MRSA-II D, isolate M06/0075 (from [12])	Human	1	+	+	-