



Ariane Deplano*, Frédéric Laurent**, François Vandenesch**, Anne Tristan**, Olivier Denis* and the EQA - ESGS-Group#

* Centre National de Référence – *S. aureus*, Hôpital Erasme – Université Libre de Bruxelles (ULB), Brussels, Belgium. ** Centre National de Référence des Staphylocoques, Hospices civils de Lyon, France. # EQA - ESGS-group, Henrik Westh, Jesper Boye Nielsen; Anders Rhod Larsen, Angela Kearns, Franziska Layer, Berit Schulte, Christiane Wolz, Iris Spiliopoulou, Joanna Empel, Waleria Hryniewicz, Artur Sabat, Alex Friedrich, Herminia de Lencastre, Nuno A Faria

Objectives

To assess ability of labs to perform by using their own phenotypic and genotypic methods:

- Identification
- Antimicrobial susceptibility testing
- Toxin, virulence, resistance gene detection
- Resistance gene detection
- Typing

Participants

11 laboratories from 9 countries including Belgium, Denmark (n=2), England, France, Germany (n=2), Greece, Poland, Portugal and The Netherlands

8 National Reference Centres (NRC)

- Basic panel: three laboratories
- Extended panel: eight laboratories

Table 1: Characteristics of the panel

Sample code	Species	MIC (mg/L) to			Resistant to	Presence of resistance gene					
		oxa	cefox	mupi		mecA	mecC	mupA	TET	MLS	AMINO
SA-01	<i>S. aureus</i>	≤ 2	≤ 4	≤ 1	ery, clin	-	-	-	-	ermA	-
SA-02	<i>S. aureus</i>	≤ 2	≤ 4	≤ 1	tet	-	-	-	tetK	-	-
SA-03	<i>S. aureus</i>	≥ 4	≥ 8	≤ 1	oxa, cef, kan, cip, ery	+	-	-	-	-	aph3
SA-04	<i>S. aureus</i>	≥ 4	≥ 8	≤ 1	oxa, cef	-	+	-	-	-	-
SA-05	<i>S. aureus</i>	≥ 4	≥ 8	≤ 1	oxa, cef, fus	+	-	-	-	-	-

Sample code	Presence of toxin gene	arcA (ACME)			spa type	SCCmec	MLST Profile	ST
		arcA	etd	seh				
SA-01	PVL	-	-	-	t034	-	3-35-167-2-20-26-39	1232
SA-02	eta, etb	-	-	-	t159	-	6-441-6-2-7-14-5	3048 (SLV ST121)
SA-03	PVL	+	-	-	t008	IV	3-3-1-14-4-3	8
SA-04	-	-	-	-	t843	XI	6-193-45-2-7-58-52	1245
SA-05	TSST-1, sec, sed	-	-	-	t002	I	1-4-1-4-12-1-10	5

Programme details

5 well-characterized *S. aureus* strains

Choice between 2 panels

- **Basic panel**
Identification,
MIC to oxacillin, cefoxitin and mupirocin
Gene detection for *mecA*, *mecC*, *mupA*, PVL, TSST-1, eta, etb
Molecular typing by one method
- **Extended panel**
Basic panel +
Antimicrobial susceptibility testing (n=16),
Resistance genes to tetracyclines (TET), aminoglycosides (AMINO) and macrolides-lincosamide (MLS)
Gene detection for *arcA*, *etd*, *seh*
SCCmec typing

Table 2: Methods used

	Maldi	Etest	broth/ agar dilution	PCR	microarray	WGS	sequencing	Ridom StaphType	BioNumerics
Identification	6			6	1	1	1		
Antimicrobial susceptibility		9	3						
Virulence/ resistance genes				8	3	3			
Molecular typing									
<i>arcA</i> , <i>seh</i> , <i>etd</i>				4	3	2			
SCCmec				6	3	2			
<i>spa</i> typing						1	9	9	2
MLST						2	7		

Analysis of concordance with expected results

- **Identification:**
 - 100 % concordance
- **Antimicrobial susceptibility testing**
 - **Phenotypic:** 100 %, except for MIC to oxa of SA-04 (*mecC*+ and LLR oxa) in 4 labs using E-test (n=3) or Sensititre (n=1)
 - **Genotypic:** 100% , except for MLS and aminoglycoside resistance in 3 labs using PCR (n=2) and WGS (n=1)
- **Virulence**
 - **Toxin:** 100% for PVL but discrepancy for TSST-1 and exfoliatine A/B in 2 labs using WGS
 - **Enterotoxin:** discrepancy observed for *sea*, *sec* and *sed* in 3 labs using WGS (n=2) or PCR (n=1)
- **Molecular typing**
 - 100% concordance for *spa* typing, SCCmec and MLST analysis
 - Note that 3 labs didn't detected the SLV od ST121 (new allele 441 for *aroE*)

Conclusions

- **Complete concordance with expected results for:**
 - Identification, MIC cefoxitin and mupirocin
 - Detection of *mecA*, *mecC*, *mupA* and PVL genes
 - Molecular typing
- **mecC positive *S. aureus***
 - Poor phenotypic detection of LLR to oxacillin
- **WGS not efficient**
 - To detect genes encoding toxins and enterotoxins