

Supporting information for Etz *et al.* (May 7, 2002) *Proc. Natl. Acad. Sci. USA*,

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Table 1. Immunogenic proteins frequently identified by bacterial surface display

ORF number (TIGR)	Name, putative function or homology	Localization motif*	Gene distribution: positive/total [†]	Hits per ORF [†]				Location of antigenic region	Reactivity with sera: positive/total (MACS recovery) [§]
				LamB		FhuA			
				Patient	Healthy	Patient	Healthy		
SA0095	Protein A	SP, LPXTG	30/30	63	–	53	36	aa 1–48 aa 47–143 aa 219–285 aa 345–424	20/28 1/1 17/28 37/71
SA0209	Coagulase	SP	30/30	–	–	–	26	aa 438–516 aa 505–570 aa 569–619	3/5 1/7 2/7
SA0317	Lipase	SP	Variable (2)	45	–	42	3	aa 48–136 aa 128–172 aa 201–258	6/13 10/28 (100%) 7/28
SA0470	Putative Exotoxin 2	SP	29/30	–	3	–	101	aa 1–80 aa 49–116 aa 98–190	6/7 25/28 13/17
SA0507	LysM domain protein	SP	30/30	–	–	2	1	aa 21–118	50/52
SA0723 [¶]	LysM domain protein, homology with SA2581	SP	30/30	–	–	58	18	aa 45–105 aa 103–166 aa 66–153	1/1 13/27 47/51
SA0858	Empbp, extracellular matrix and plasma-binding protein	SP	21/30	27	2	50	15	aa 22–56 aa 23–99 aa 97–115 aa 233–250 aa 245–265	58/71 59/59 1/1 ND ND
SA1062	Atl, autolysin	SP	29/30	47	5	22	61	aa 6–66 aa 65–124 aa 590–604	5/16 15/28 39/71
SA1472	Pathogenicity protein	SP		–	–	10	30	aa 8511–8640	5/26 (100%)
SA1781	LPXTGp5	SP, LPXTG	30/30	22	3	1	–	aa 37–49 aa 63–77 aa 274–334	1/1 12/70 4/28
SA2004	Putative leukocidin F precursor			–	–	9	–	aa 158–220	7/27 (100%)
SA2006	Putative aerolysin	SP	30/30	3	4	12	36	aa 31–61 aa 58–74	19/27 49/71
SA2019	Putative SdrH	SP	30/30	7	9	–	–	aa 122–139 aa 164–182	ND 39/71
SA2291	Homology with SA2581	SP	30/30	58	42	224	169	aa 137–237 aa 250–267	13/27 10/71
SA2295	Homology with SA2581	SP		4	–	5	–	aa 38–52 aa 66–114	ND 4/28
SA2418	Sbi, IgG-binding protein	SP	97% (3)	11	23	1	–	aa 208–287 aa 286–314	38/46 11/71
SA2505	LPXTGp4	SP, LPXTG		–	18	4	3	aa 491–585 aa 631–713 aa 702–758	1/1 1/1 15/28
SA2509	FnbpB, Fibronectin binding	SP	100% (4)	–	2	2	5	aa 693–769 aa 775–814	26/26 1/1
SA2511	FnbpA, Fibronectin binding	SP	100% (4)	–	2	4	4	aa 710–787 aa 855–975	19/25 ND
SA2581	Staphyloxanthin biosynthesis protein, homolog of <i>Staphylococcus epidermidis</i> SsaA	SP	26/30	51	43	130	13	aa 916–983 aa 7–87 aa 133–242	15/28 1/1 5/27 (100%)

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				LamB		FhuA			
				Patient	Healthy	Patient	Healthy		
SA2584	IsaA	SP	30/30	69	2	8	3	aa 67–116 aa 98–184 aa 182–225	1/1 20/26 34/71
SA2659 SAA0001	Aureolysin repC	SP	100% (5)	– –	– 6	1 –	6 18	aa 83–156 aa 9–42 aa 158–174	13/27 1/1 1/1

Screens of two libraries with two serum pools each are summarized: LSA250 library in FhuA with patient serum P1 (655 clones analyzed) and with serum from healthy individuals N1 (680); LSA50 library in LamB with patient sera P1 (498) and with serum from healthy individuals N2 (900). Only a selection of frequently identified antigens is shown. *, Motifs characteristic of secreted or surface exposed proteins: SP, signal peptide; LPXTG, sortase anchoring motif; †, PCR analysis was performed with oligonucleotides designed to amplify approximately 500- to 1,000-bp fragments covering at least one antigenic epitope. Twenty-four *Staphylococcus aureus* strains were isolated from patients and three strains from healthy individuals and typed according to restriction fragment length polymorphism (RFLP) of the *spa* and *coa* genes (1). The three laboratory strains, COL, Newman, and 8325-4, were used as controls. PCR products of variable size were obtained for the SA2019, SA0095, and SA0209 genes. Sequencing of the PCR products revealed that the size difference was due to a variable copy number of a repetitive DNA sequence in these three genes. ‡, The number of clones identified per ORF is given for the individual screens. Representative clones were analyzed further. §, One epitope of the indicated region was chosen for Western blot analysis with individual sera. Several epitopes identified frequently were not or were only very weakly reactive in Western blot analysis indicating the presence of conformational epitopes. These clones were therefore tested in MACS experiments where the rate of specific recovery with biotinylated patient serum P1 is expressed as percentage of the input

(shown in brackets). ¶, Identified 18 times of 33 in a FhuA prescreen with serum from healthy individuals N1, and was therefore eliminated from the library prior to screen FhuA with N1. ND, not determined; MACS, magnetic cell sorting.

References

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