

Panton-Valentine Leukocidin Genes in *Staphylococcus aureus*

To the Editor: The pathogenicity of *Staphylococcus aureus* depends on various bacterial surface components and extracellular proteins. However, the precise role of single virulence determinants in relation to infection is hard to establish. The frequent recovery of staphylococcal isolates that produce leukocidal toxins from patients with deep skin and soft tissue infections, particularly furunculosis, cutaneous abscesses, and severe necrotizing pneumonia, suggests that the Panton-Valentine leukocidin (PVL) is 1 such virulence factor that has a major role in pathogenicity (1–3).

In 1932, Panton and Valentine described PVL as a virulence factor belonging to the family of synergohyminotropic toxins (4). These toxins form pores in the membrane of host defense cells by synergistic action of 2 secretory proteins, designated LukS-PV and LukF-PV, which are encoded by 2 cotranscribed genes of a prophage integrated in the *S. aureus* chromosome (5). PVL is mostly associated with community-acquired methicillin-resistant *S. aureus* (MRSA) infections and distinguish-

able from nosocomial MRSA by non-multidrug resistance and carriage of the type IV staphylococcal chromosome cassette element (SCC_{mec} type IV) (6,7).

Despite the presumed importance of PVL as a virulence factor, few data are available on its prevalence among *S. aureus* isolates from the nares of healthy persons compared with strains isolated from infections. This lack of data led us to investigate the frequency of PVL gene-positive *S. aureus* strains obtained from the nares of healthy carriers in the community. For this purpose, a single polymerase chain reaction method was used to detect both *lukS-PV* and *lukF-PV* genes (2).

In a previous study, the population structure of *S. aureus*, isolated from the nares of healthy persons in the Rotterdam area, the Netherlands, was elucidated (8). Strains were obtained from healthy children (<19 years) and elderly persons (>55 years). Invasive strains (blood culture, skin and soft tissue infections, and impetigo isolates) were included in this study (Table). All carriage and clinical isolates (n = 1,033) were *mecA* negative. We used the same strain collection to study the PVL prevalence in carriage and invasive isolates of *S. aureus* from a single geographic region.

Five PVL-positive *S. aureus* strains (0.6%) were found in the car-

riage group (n = 829), and 3 (2.1%) of 146 blood-culture isolates carried the PVL gene (Table). This finding is in agreement with previously reported low PVL prevalences by Prevost et al. (0% in 31 carriage isolates and 1.4% in 69 blood-culture isolates) and Von Eiff et al. (1.4% in 210 carriage isolates and 0.9% in 219 blood-culture isolates) (9,10). However, a higher prevalence of PVL (38.9%) was found in *S. aureus* strains causing abscesses and arthritis (Fisher exact test, p <0.0001) (8). This finding is also in agreement with the proposed involvement of PVL in severe and invasive (soft tissue) staphylococcal infections (1–3). No significant differences were found in the presence of PVL when carriage isolates were compared with invasive blood-culture isolates. PVL was found in each major genomic amplified fragment length polymorphism (AFLP) cluster, indicating that PVL has been introduced in distinct phylogenetic subpopulations of *S. aureus* (online Figure; available from <http://www.cdc.gov/ncidod/EID/vol112no07/05-0865-G.htm>). Multilocus sequence typing analysis of a subset of the strain collection showed that the 15 PVL-positive strains were within clonal complex (CC) 30 (n = 7), CC 121 (n = 3), CC 1 (n = 2), CC 8 (n = 1), CC 22 (n = 1), and CC 45 (n = 1) (Table) (8). Although PVL was found among several staphylococcal

Table. Panton-Valentine Leukocidin (PVL) distribution among carriage and invasive isolates per genetic cluster of *Staphylococcus aureus*

	Amplified fragment length polymorphism cluster					Total (N = 1,033)
	I (n = 462)	II (n = 261)	III (n = 208)	IVa (n = 62)	IVb (n = 40)	
	PVL positive, n (%)					
Carriage isolates (n = 829)	1	1	1	0	2	5 (0.6)*
Bacteremia isolates (n = 146)	1	1	0	0	1	3 (2.1)†
Soft tissue infection isolates (n = 18)	1	5	0	1	0	7 (38.9)‡
Impetigo isolates (n = 40)	0	0	0	0	0	0 (0.0)
Total (N = 1,033)	3 (0.6)§	7 (2.7)	1 (0.5)¶	1 (1.6)	3 (7.5)#	15 (1.5)
MLST** data of PVL-positive isolates	CC 1, n = 2 CC 8, n = 1	CC 30, n = 7	CC 45, n = 1	CC 22, n = 1	CC 121, n = 3	

‡ versus * Fisher exact test (2-sided); p <0.0001.

† versus ‡ Fisher exact test (2-sided); p <0.0001.

versus § Fisher exact test (2-sided); p = 0.0079.

¶ versus ¶ Fisher exact test (2-sided); p = 0.0140.

**MLST, multilocus sequence typing; CC, clonal complex.

genotypes, it was slightly overrepresented in AFLP cluster IVb (CC 121) compared with major clusters I and III. Whether the prevalence of PVL in carriage- and blood-culture isolates is higher and differs among distinct genetic clusters of *S. aureus* in countries with endemic CA-MRSA has to be investigated further.

In conclusion, we have shown that the PVL-encoding phage has entered distinct staphylococcal lineages, although its prevalence differs per clonal group. PVL is associated with skin and soft tissue infections but not with bacteremia, which suggests that PVL is not likely to be involved in the pathogenesis of bacteremia. Infections caused by PVL-positive *S. aureus* strains have been documented since the 1930s. Expansion and increased incidence of such infections, however, are more recent, and further epidemiologic studies for tracking this phenomenon are still warranted.

**Damian C. Melles,*
Willem B. van Leeuwen,*
Hélène A.M. Boelens,*
Justine K. Peeters,*
Henri A. Verbrugh,*
and Alex van Belkum***

*University Medical Center Rotterdam, Rotterdam, the Netherlands

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Address for correspondence: Damian C. Melles, Erasmus MC, University Medical Center Rotterdam, Department of Medical Microbiology & Infectious Diseases, Rm L-313, Dr Molewaterplein 40, 3015 GD Rotterdam, the Netherlands; email: d.melles@erasmusmc.nl



Small Anellovirus in Hepatitis C Patients and Healthy Controls

To the Editor: Torquetenovirus (TTV) and torquetenominivirus (TTMV) are characterized by a small, negative-sense, circular, single-stranded DNA genome and by an extraordinary ability to produce chronic plasma viremia. Indeed, >80% of humans harbor variably high viral loads of TTV, TTMV, or both, in plasma, regardless of geographic provenance, age, sex, and health conditions (1). Currently, TTV and TTMV are classified as distinct species in the floating (although closely linked to the family *Circoviridae*) genus *Anellovirus*, but their extreme genetic heterogeneity and some distinctive features in genomic organization have led some to suggest that they should be classified as an independent family (2,3). Most recently, after examining serum specimens from patients with symptoms of an acute viral infection by using DNase sequence-independent single-primer amplification, Jones et al. (4) identified, among other viruses, 2 novel TTV- and TTMV-like agents. Because of their even smaller genomes (≈ 2.4 and 2.6 kb vs. 3.6–3.8 kb for TTV and 2.8–2.9 kb for TTMV), these agents were named small anelloviruses (SAVs).

Because tissue culture and serologic methods are not yet available, diagnosis of anellovirus infection relies exclusively on viral DNA detection. We tested 55 Italian hepatitis C patients (mean age 56 ± 14 years, male/female ratio 30/25, 53 TTV positive) and, for comparison, 35 healthy donors (mean age 36 ± 12 years, male/female ratio 17/18, 33 TTV positive) for SAV in plasma by using the polymerase chain reaction (PCR) primers described by Jones et al. (4), followed by direct amplicon