# Staphylococcus aureus Causing Tropical Pyomyositis, Amazon Basin, Peru

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We studied 12 *Staphylococcus aureus* isolates causing tropical pyomyositis in the Amazon Basin of Peru. All isolates were methicillin-susceptible; 11 carried Panton-Valentine leukocidin–encoding genes, and 5 belonged to multilocus sequence type 25 and possessed an extensive set of enterotoxins. Our findings suggest sequence type 25 is circulating in tropical areas of South America.

Pyomyositis is an acute bacterial infection characterized by suppuration within large skeletal muscles manifesting as single or multiple abscesses. Its exact pathogenesis is unknown but is thought to occur through bacteremic seeding (1). The infection is often seen in tropical countries, hence the name tropical pyomyositis, and usually occurs in young, otherwise healthy persons. In addition, the infection is increasingly reported from temperate regions in patients receiving immunosuppressive therapy or with HIV infection (1,2). The most common bacterial causes of tropical pyomyositis are Staphylococcus aureus (90% in tropical areas, 75% in temperate zones) and group A streptococcus (1%-5%); less common causes are group B, C, and G streptococcus, pneumococcus, Haemophilus spp., and gram-negative bacilli (2). Furthermore, the increasing incidence of pyomyositis in temperate regions has been correlated with the emergence and spread of community-associated (CA)-methicillinresistant S. aureus (MRSA) clones (3). These CA-MRSA clones usually produce Panton-Valentine leukocidin (PVL), a pore-forming toxin encoded by 2 genes, lukF-PV and lukS-PV (4).

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Few studies have been done on the genetic characteristics of *S. aureus* that cause tropical pyomyositis. To help fill this void, we determined the molecular characteristics of *S. aureus* isolates causing tropical pyomyositis in the Amazon Basin of Peru.

## The Study

We analyzed 12 *S. aureus* isolates from patients with tropical pyomyositis. Of the 12 isolates, 10 were obtained from patients hospitalized in a 120-bed public hospital located in Iquitos, the largest city in the Amazon Basin of Peru (160,000 inhabitants). A retrospective chart review showed that 38 patients were hospitalized with tropical pyomyositis during 2009–2010; these patients represented 0.9% of 4,445 hospital admissions. The 10 isolates from hospitalized patients came from these patients; the 2 other isolates were obtained in 2005 from 2 persons hospitalized in Yurimaguas, a city 388 km southeast of Iquitos (63,000 inhabitants). We obtained clinical data by reviewing patients' charts.

Isolates were analyzed at the Instituto de Medicina Tropical Alexander von Humboldt in Lima, Peru. Colonies were identified as S. aureus by Gram staining and by positive reactions for catalase, DNAse, and tube coagulase tests. We screened for oxacillin resistance by using the cefoxitin (30  $\mu$ g) disk diffusion test, and we used disk diffusion to assess susceptibilities to clindamycin, erythromycin, gentamicin, ciprofloxacin, rifampin, and trimethoprim-sulphamethoxazole (5).

Detection of 16S rRNA, mecA, and nuc genes by multiplex PCR was performed at the Centre National de Référence S. aureus, Erasme Hospital, Brussels, Belgium (6). We used PCR to detect the presence of the PVL- and toxic shock syndrome toxin 1-encoding genes (lukS-lukF PV and tst, respectively), exfoliatin A- and B-encoding genes (eta and etb, respectively) (7), and enterotoxin genes (sea, seb, sec, sed, see, seg, seh, sei, sej, sek, sem, sen, seo, sep, seq, ser) as described by Omoe et al. (8). We determined spa type as described (9), and we performed multilocus sequence typing on 1 randomly selected isolate of each spa type (10).

Demographic characteristics of the patients are shown in the Table. Of the 12 patients, 9 (75%) were <5 years of age, similar to age distributions recorded in other studies (2). No patient revealed a history of chronic disease. Two groups of muscles were involved in 2 patients, and 10 patients had fever. All patients had local edema and pain, needed surgical drainage, and had received antimicrobial drugs (oxacillin and/or clindamycin) before samples were obtained.

Of the 12 *S. aureus* isolates we studied, 11 (92%) carried PVL genes; PVL genes were also reported in 12% of clinical MSSA isolates recovered in hospitals in Lima (11). In the United States and France, PVL has been detected in

Table. Characteristics of patients with tropical pyomyositis caused by methicillin-susceptible *Staphylococcus aureus*, Amazon Basin, Peru\*

	Year patient	Body site	S. aureus isolate							
Patient			spa	Multilocus	Gene					
age	hospitalized	involved	type	ST†	mecA	eta	etb	lukS-lukF	tst	Enterotoxin genes
12 mo	2005	Lower limb	t6465	ST121	_	_	_	+	_	sei, seg
3 y	2005	Lower limb	t6465	ST121	_	_	_	+	_	sei, seg
6 mo	2009	Upper limb	t701	ST6	_	_	_	+	_	sea
12 y	2009	Thorax, lower limb	t078	ST25	-	-	_	+	-	seb, sed, sei, sej, seg, sem, sen, seo, ser
24 mo	2009	Lower limb	t078	ST25	-	-	=	+	_	seb, sed, sei, sej, seg, sem, sen, seo, ser
45 y	2009	Thorax	t1778	ST1	_	_	_	+	_	seh
3 y	2009	Abdomen, upper limb	t701	ST6	-	-	-	+	-	sea
28 d	2010	Lumbar	t164	ST20	_	_	_	-	_	seg, sei, sem, sen, seo
3 y	2010	Lower limb	t078	ST25	-	-	-	+	-	seb, sed, sei, sej, seg, sem, sen, seo, ser
26 y	2010	Abdomen	t078	ST25	-	-	_	+	-	seb, sed, sei, sej, seg, sem, sen, seo, ser
4 y	2010	ND	t1778	ST1	_	_	_	+	_	seh
18 mo	2010	Thorax	t078	ST25	-	-	_	+	-	seb, sed, sei, sej, seg, sem, sen, seo, ser

<sup>\*</sup>ST, sequence type; -, negative; +, positive; ND, no data.

20% and 10%, respectively, of CA-MSSA soft tissue infections (3,12). S. aureus carrying PVL has received more attention by public health officials since the emergence, in the United States and other countries, of skin and soft tissue infections caused by the PVL-positive CA-MRSA clone USA300. A recent publication from the United States noted that 69% of MRSA isolates causing skin and soft tissue infections carried PVL (3).

None of the isolates described in this study carried genes associated with the toxic shock syndrome toxin (tst) or the staphylococcal scalded skin syndrome (eta, etb), but all possessed several enterotoxin genes. The presence of toxin genes was strongly linked to spa type (Table). In particular, isolates belonging to spa type t078 (sequence type [ST] 25) harbored the egc operon (coding genes seg, sei, sem, sen, seo), coding genes sed, sej and ser (probably carried on the same plasmid) (13), and seb. Along with toxic shock syndrome toxin 1, seb is one of the most powerful staphylococcal superantigens.

Of particular interest in our study is the high proportion of isolates belonging to ST25. This ST has been detected in bovine and human isolates in the United Kingdom (14), and an ST25 PVL-positive MSSA was detected in a patient in Brazil with life-threatening sepsis with pneumonia and myositis (15). ST25 PVL-positive isolates are thought to be rare in Peru: in 2008–2009, only 6 (3.6%) of 169 MSSA blood isolates from hospitalized patients in Lima (non-tropical area of Peru) harbored spa type t078 or related spa types (C. Garcia, unpub. data). Our findings raise the hypothesis that ST25 is circulating in the tropical areas of South America.

This study has several limitations. First, tropical pyomyositis cases from the referring hospitals were defined by the patients' physician, and isolates from all patients were not assessed. Second, because this was a retrospective study, many variables were not recorded, including history of blunt trauma and duration of antimicrobial drug therapy. However, given that studies of the genetic characteristics of *S. aureus* that cause tropical pyomyositis are lacking and given the striking presence of PVL genes among the isolates in our study, our findings add to the insights about the pathogenesis of this acute suppurative infection.

### Conclusions

This study describes a high rate of PVL encoding genes among *S. aureus* causing tropical pyomyositis in the Amazon Basin of Peru. Further investigation in areas geographically different from the Amazon Basin should be done to confirm the association of PVL and other toxins in the pathogenesis of tropical pyomyositis.

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<sup>†</sup>Boldface indicates isolates that were randomly selected by spa type for multilocus sequence typing (MLST). The other isolates were not analyzed by MLST. Results for those isolates were extrapolated from results for the analyzed isolates of the same spa type.

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