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Involvement of Staphylococcal Enterotoxins in Nonmenstrual Toxic Shock Syndrome

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Adequate evidence is available to show that the major toxin responsible for toxic shock syndrome (TSS) is TSS toxin 1 (TSST-1). More than 90% of the staphylococcal strains isolated from TSS patients produce this toxin. However, approximately 60% of these strains produce one or more of the staphylococcal enterotoxins, with a number of them producing only enterotoxin, primarily enterotoxin B. Of 55 staphylococcal strains isolated from nonmenstrual cases, 46 produced TSST-1; 42 produced one of the enterotoxins, including 8 that produced only enterotoxin B. The fact that the enterotoxins can produce in monkeys many signs and symptoms similar to those observed in TSS in humans implicates them as the cause of some cases of TSS.

Toxic shock syndrome (TSS) was first described by Todd et al. in 1978 (13) as a disease of children. Two years later, TSS received medical and public recognition as a widespread clinical entity affecting young, healthy menstruating women, especially women who used tampons (12). Vaginal isolates of *Staphylococcus aureus* (coagulase and thermonuclease positive) from patients with TSS were shown to produce a new toxin, independently identified by Bergdoll et al. (2) and Schlievert et al. (11). With the heightened awareness of TSS, nonmenstrually associated cases have been recognized with increasing frequency (10). Nonmenstrual and menstrual cases are similar in clinical and laboratory findings, but differ in epidemiology. Nonmenstrual cases have occurred in males and females of all ages and races, whereas menstrual cases are seen predominately in white women.

Bergdoll et al. (3) showed that the toxin, TSS toxin 1 (TSST-1) (4), associated with the staphylococci isolated from TSS patients was produced by 130 (91.5%) of 142 TSS *S. aureus* strains; of 22 of these strains that were isolated from nonmenstrual cases, 20 (90.9%) produced TSST-1. Schlievert et al. (11) reported that 28 of 28 TSS *S. aureus* strains isolated from menstrual cases produced TSST-1. Garbe et al. (7) found that 44 (91.7%) of 48 *S. aureus* strains isolated from menstrual TSS cases produced TSST-1, but only 9 (56.3%) of 16 strains from nonmenstrual TSS cases produced TSST-1. Reingold et al. (10) reported that 12 (85.7%) of 14 isolates from nonmenstrual cases produced TSST-1. A total of 223 (89.9%) of the 248 strains produced TSST-1. The involvement of TSST-1 in TSS is unquestioned; it has been shown to produce the signs and symptoms of TSS, except desquamation, when injected intravenously into baboons (9). However, the fact that 85 (59.8%) of the 142 TSS isolates examined by Bergdoll et al. (3) produced one or more of the staphylococcal enterotoxins, with 7 (8.2%) not producing TSST-1, indicated that the enterotoxins also may be involved in TSS, particularly in those cases where the isolates did not produce TSST-1. Experiments have shown that the intravenous injection of the enterotoxins into rhesus monkeys can produce many signs and symptoms similar to those observed in TSS in humans: fever, hypotension, vomiting, diarrhea, elevation of blood urea nitrogen, increase in serum creatinine, decrease in urine output, increase in glutamic oxalacetic transaminase, thrombocytopenia, hyperfibrinogenemia, initial leukopenia followed by a neutrophilic leukocytosis, pulmonary edema, increase in heart rate, pooling of the blood in vascular beds and evidence of endothelial cell degeneration, gradual decrease in serum proteins, shock, and death (1).

We routinely examine all TSS *S. aureus* strains provided to us for the production of the enterotoxins as well as for TSST-1 because we feel that this information may be important in resolving some of the cases of TSS. In light of the Garbe et al. report (7) that some nonmenstrual cases were due to a toxin(s) other than TSST-1, we analyzed the data we had accumulated on the production of toxins by *S. aureus* strains isolated from nonmenstrual TSS cases; the result of this analysis is reported here.

We examined isolates from 55 people who had either confirmed (38) or probable (17) cases of nonmenstrual TSS, as described by the Centers for Disease Control criteria (6). Patients who had TSS postpartum or due to vaginal infection were excluded from the study. The cases occurred between December 1979 and October 1983. The patients included 23 (41.8%) females and 32 (59.2%) males. There was no difference with regard to sex for the mean or median age: 21 and 15 years, with a range of 1 day to 67 years. The majority of the cases were from the Midwest, predominately Wisconsin, Michigan, Iowa, and Illinois. Three cases were from South Africa (8), and one was from Iceland. Five cases were fatal. Of the 55 *S. aureus* isolates, 20 (36.4%) were from skin lesions: nonsurgical wounds, ingrown nails, cellulitis, and burns. Of the remaining 35, 12 (21.8%) were from abscesses, 10 (18.6%) were from surgical wounds, 5 (9.1%) were from blood from children, 4 (7.3%) were from bone or aspirate in cases of osteomyelitis or septic arthritis in children, 3 (5.4%) were from sputum from pneumonia, and 1 (1.8%) was from an infant with tracheitis.

The 55 *S. aureus* isolates were grown and tested for TSST-1 and staphylococcal enterotoxin A (SEA), SEB, SEC, SED, and SEE by the membrane-over-agar and optimum sensitivity plate method (5) (Table 1). A total of 46 (83.6%) of the 55 isolates produced TSST-1: 12 (25.5%) alone, 21 (46.8%) with SEA, and 13 (27.7%) with SEC. Eight of the isolates that did not produce TSST-1 did produce SEB, one of the enterotoxins used in the monkey studies

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TABLE 1. Production of TSST-1 and the staphylococcal enterotoxins by *S. aureus* isolated from nonmenstrual TSS cases

Infection	No. of strains producing toxin:				
	TSST-1	TSST-1 + SEA	TSST-1 + SEC	SEB	None
Skin lesions	6	8	3	2	1
Abscesses	4	4	3	1	0
Surgical wounds	1	7	1	1	0
Blood	1	0	3	1	0
Osteomyelitis/ septic arthritis	0	1	0	3	0
Pneumonia	0	0	3	0	0
Tracheitis	0	1	0	0	0

mentioned above (1). To date we have not examined a strain that produced both SEB and TSST-1. Acute and convalescent sera were available for analysis from five of these eight patients. One patient had no antibodies in either the acute or convalescent sera to either TSST-1 or SEB, three showed an increase in SEB antibodies with no change in a positive TSST-1 antibody titer, and one showed an increase in titer to both TSST-1 and SEB. Involvement of TSST-1 is indicated in this last patient, and no firm conclusion may be made about the possible involvement of TSST-1 in the patient with no antibodies to either toxin. The antibody titers of the remaining three patients implicate SEB, especially because the TSST-1 antibody titers were greater than 1:100.

Analysis in our laboratory of the seven TSS *S. aureus* strains from nonmenstrual cases reported by Garbe et al. (7) to be TSST-1 negative showed five of them to be enterotoxin positive with four strains producing SEB. It is true that SEB was isolated a number of years (3) before TSS was identified as a specific disease (13); however, after TSS was recognized it became apparent that illnesses similar to TSS resulting from staphylococcal infections had occurred for many years.

The major evidence that any toxin is involved in a particular disease is through animal studies, as it is not possible to conduct experiments involving humans. The results of animal experiments with SEB showed that this toxin produces many of the signs and symptoms similar to those of TSS (1), indicating that if the toxin is produced in humans during an infection, TSS may result. The increase in antibody titers to SEB in several patients during convalescence indicated that SEB was produced during the illnesses.

Roughly 3 to 5% of staphylococcal strains isolated from TSS patients are negative for any of the toxins. It is always possible that the strain actually involved in the disease may not have been isolated, because we have found that in cases where multiple isolates were received not all produced toxin (5). There is no evidence that these strains produce unidentified toxins, but one cannot dismiss the possibility that other toxins do exist. Even though other toxins may exist, evidence that SEB is involved in some cases of TSS is still valid.

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