

# Isolation of an Additional Capsular-Type Strain of *Staphylococcus aureus* by the Serum-Soft Agar Technique

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An additional capsular-type strain of *Staphylococcus aureus* was isolated by the serum-soft agar technique and designated as capsule type D.

Previously (1), I reported on three serologically different capsular-type *Staphylococcus aureus* strains. These strains were isolated from humans by the serum-soft agar (SSA) technique and were designated as types A, B, and C. Recently, an encapsulated *S. aureus* strain (NS68D), isolated from the throat of a patient at the Nippon Medical School Hospital, Tokyo, Japan, was

those noted previously (3). Determination of its growth type in SSA, bacteriophage- and serotyping, and the capsule staining method were performed as described elsewhere (3). The capsule-typing method, the procedure for the adsorption of converting activity, examination for conversion of diffuse to compact growth type in SSA containing antisera, and the procedure for preparation

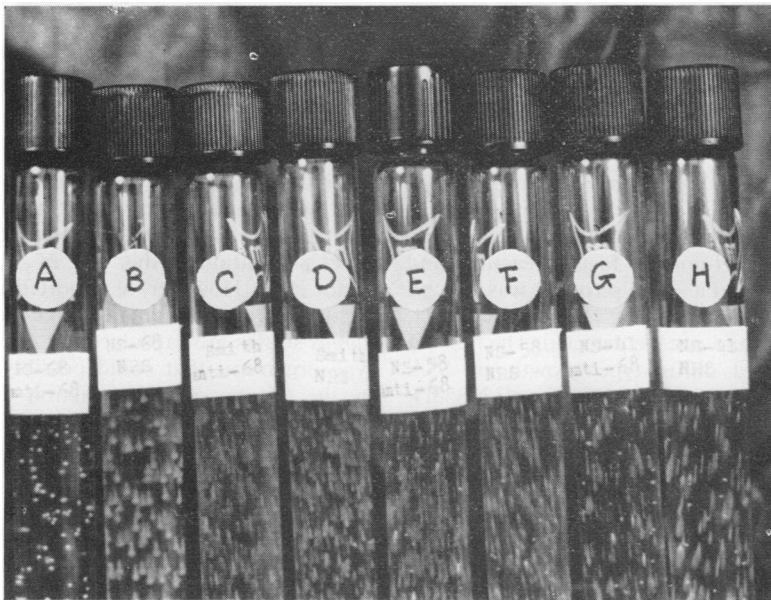


FIG. 1. Colonial morphologies of the strains *Smith diffuse*, NS58D, NS41D, and NS68D in regular serum-soft agar containing normal rabbit sera: (B) NS68D, (D) *Smith diffuse*, (F) NS58D, and (H) NS41D; and containing anti-NS68D sera: (A) NS68D, (C) *Smith diffuse*, (E) NS58D, and (G) NS41D.

found to possess a capsule serologically different from those noted above. This paper is concerned with these investigations.

Strain NS68D is a nonmotile, catalase-positive, gram-positive coccus. Procedures for the determination of its glucose utilization, coagulase, deoxyribonuclease, mannitol fermentation, acid phosphatase, and clumping factor activities were

of antisera were also followed as described previously (1).

Strain NS68D showed positive reactions for glucose oxidation/fermentation, free coagulase, and deoxyribonuclease and for mannitol fermentation and acid phosphatase. It gave a negative clumping factor reaction, was nontypable both for bacteriophage and serotyping, and



FIG. 2. Colonial morphologies of the strain NS68D in regular serum-soft agar containing (A) anti-NS68D serum untreated, (B) normal rabbit serum, (C) anti-NS68D serum absorbed with NS68D organisms, (D) anti-NS68D serum absorbed with *Smith diffuse* organisms, (E) anti-NS68D serum absorbed with NS58D organisms, and (F) anti-NS68D serum absorbed with NS41D organisms.

showed diffuse-type growth in SSA; the capsule was demonstrable by a light microscope. A compact variant, isolated from a diffuse-type culture, gave a positive clumping factor reaction and was phage nontypable. The serotype was b h<sub>2</sub> m 263-2 by Oeding and I, 10, 11, 17 by Pillet, and a capsule could not be detected. The NS68D strain represented diffuse-type growth in SSA containing strains anti-Smith diffuse, anti-NS58D, or anti-NS41D. The converting activity of these antisera remained unaffected by heterologous strains but was absorbed with homologous organisms. Conversely, strains *Smith diffuse*, NS58D, and NS41D showed diffuse-type growth in the SSA containing anti-NS68D strain as shown in Fig. 1, and the converting activity of anti-NS68D sera was not absorbed with heterologous strains as shown in Fig. 2. Furthermore, the converting activity of anti-NS68D sera was absorbed with the diffuse type of homologous organisms but was not absorbed with counterpart compact organisms. From these results, it was evident that the strain NS68D was serologically different from strains *Smith diffuse*, NS58D, and NS41D, the capsules of which were designated as types A, B, and C, respectively. Thus, this capsule type was designated as type D. The relation of the

stability of encapsulation, which would be associated with the isolation ratio of the organisms, to the capsular type is not known; therefore, the exact population of this type of *S. aureus* strain is not clear. However, as reported previously (1, 3), the majority of human-source encapsulated *S. aureus* strains are capsular type A, *Smith diffuse*-type organisms that could possibly be subtyped by the specific capsular reaction (2). Also, this type strain was the only strain found in approximately 100 encapsulated *S. aureus* strain (*unpublished observation*). Thus, their number could be assumed as rather small in human sources. The virulence and immunological specificities of the capsule are under investigation in this laboratory, and the results will be published in future publications.

#### LITERATURE CITED

1. Yoshida, K. 1971. Demonstration of serologically different capsular types among strains of *Staphylococcus aureus* by the serum-soft agar technique. *Infect. Immunity* 3:535-539.
2. Yoshida, K., and Y. Naito. 1972. Comparison of capsular types of *Staphylococcus aureus* strains. *Infect. Immunity* 5:143-144.
3. Yoshida, K., M. R. Smith, and Y. Naito. 1970. Biological and immunological properties of encapsulated strains of *Staphylococcus aureus* from human sources. *Infect. Immunity* 2:528-532.