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Comparison of a Yellow Latex Reagent with Other Agglutination Methods for the Identification of *Staphylococcus aureus*

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A new commercial yellow latex agglutination reagent (Bacto-Staph) was compared with the slide and tube coagulase tests and three other commercial reagents for the identification of 283 *Staphylococcus aureus* and 54 non-*S. aureus* staphylococcal strains. Test sensitivities for the identification of *S. aureus* were as follows: tube coagulase, 99.6%; slide coagulase, 98.6%; Bacto-Staph, 99.6%; Staphylatex, 98.6%; Sero STAT Staph, 98.2%; and Staphyloslide, 97.5%. No false-positive reactions were observed with any of the commercial reagents.

The tube coagulase test is the procedure most relied upon by clinical laboratories to distinguish *Staphylococcus aureus* from other members of the family *Micrococcaceae* (11). However, the test takes 4 to 24 h to produce an answer (19). Therefore, many laboratories use the slide clumping factor test to screen for positive reactions, because it takes only about 1 min to perform, and then they use the tube test only for organisms which were negative by the slide test. Other slide tests which ideally would be more sensitive than the slide coagulase test have been developed for the rapid identification of *S. aureus*. A high correlation of protein A and coagulase production has been reported. In a previous study, protein A was detected in 98.9% of coagulase-positive strains tested but in only 2% of coagulase-negative strains (8). Maxim et al. described a sensitized sheep erythrocyte system and found 98.2% of the coagulase-positive strains and none of the coagulase-negative strains they tested contained protein A (13).

Several commercial modifications of these agglutination reactions have been introduced in the last few years. The Staphyloslide (BBL Microbiology Systems, Cockeysville, Md.) reagent utilizes a sensitized sheep erythrocyte system. Additionally, a combination test which simultaneously detects protein A and the clumping factor by coating latex particles with plasma was developed (6). Two commercial products that use this technique are Sero STAT Staph (Scott Laboratories, Inc., Fiskeville, R.I.) and Staphylatex (American Scientific Products, McGaw Park, Ill.). The plasma contains both fibrinogen and immunoglobulin G, and, therefore, strains producing either clumping factor or protein A or both cause agglutination. The purpose of this study was to evaluate a new commercial yellow latex reagent, Bacto-Staph (Difco Laboratories, Detroit, Mich.), which is coated with plasma, and to compare the results obtained with this reagent with those obtained with the other three commercial reagents, the slide clumping factor test, and the tube coagulase test.

The instructions of each manufacturer were followed. All four reagents were easy to use and interpret. Organisms were coded so that the technologist performing the test was unaware of their reactions in the other tests. Any discrepancies were included in the final data only after the results were confirmed by repeat testing.

Staphylococcal stock strains (91 coagulase positive and 20 coagulase negative) characterized in a previous study (3) and 224 recent clinical isolates obtained from specimens submitted to the Clinical Microbiology Laboratory of Hutzel Hospital were tested in this study.

Isolates were identified as staphylococci when the colonies were consistent with staphylococcal morphology and contained gram-positive cocci in clusters that were catalase positive and fermented glucose. Glucose fermentation was performed by the method of Facklam and Smith (7). The API Staph-Ident system (Analytab Products, Plainview, N.Y.) and novobiocin susceptibility testing (9) were used for species identification (12). The Sceptor system (Johnston Laboratories, Inc., Cockeysville, Md.) was used for MIC susceptibility testing. The procedures were performed according to manufacturer recommendations, which were previously evaluated (10). Strains were interpreted as methicillin resistant when the MIC was greater than 8 μg/ml after 48 h of incubation (15). Two control strains, *S. aureus* ATCC 25923 and *Staphylococcus epidermidis* ATCC 12228, were tested 12 times each on different days throughout the study and consistently produced positive and negative reactions, respectively, for all reagents. The reactions obtained with these control strains and with a fresh clinical isolate which produced moderate clumping in each of the agglutination reactions are shown in Fig. 1. The latter strain caused a strong reaction in the slide clumping factor test and a 4-h positive tube coagulase test.

The tube coagulase test was positive for 282 of the 283 strains (sensitivity, 99.6%) identified biochemically as *S. aureus*. The slide clumping factor test was positive for 279 strains (sensitivity, 98.6%). The Bacto-Staph test had a sensitivity of 99.6%. It detected the four strains which were negative by the clumping factor test but missed one other strain. The Staphylatex test had the next best results, detecting 279 strains for a sensitivity of 98.6%, followed by the Sero STAT Staph test, which detected 278 strains (sensitivity, 98.2%), and the Staphyloslide test, which detected 276 strains (sensitivity, 97.5%). No false-positive reactions were observed for any of the four reagents when tested against 53 coagulase-negative staphylococci (Table 1).

One of the stock strains was positive in the tube coagulase test but negative in the slide clumping factor test and with all four commercial reagents. This strain was identified biochemically as *Staphylococcus intermedius*. Coagulase-

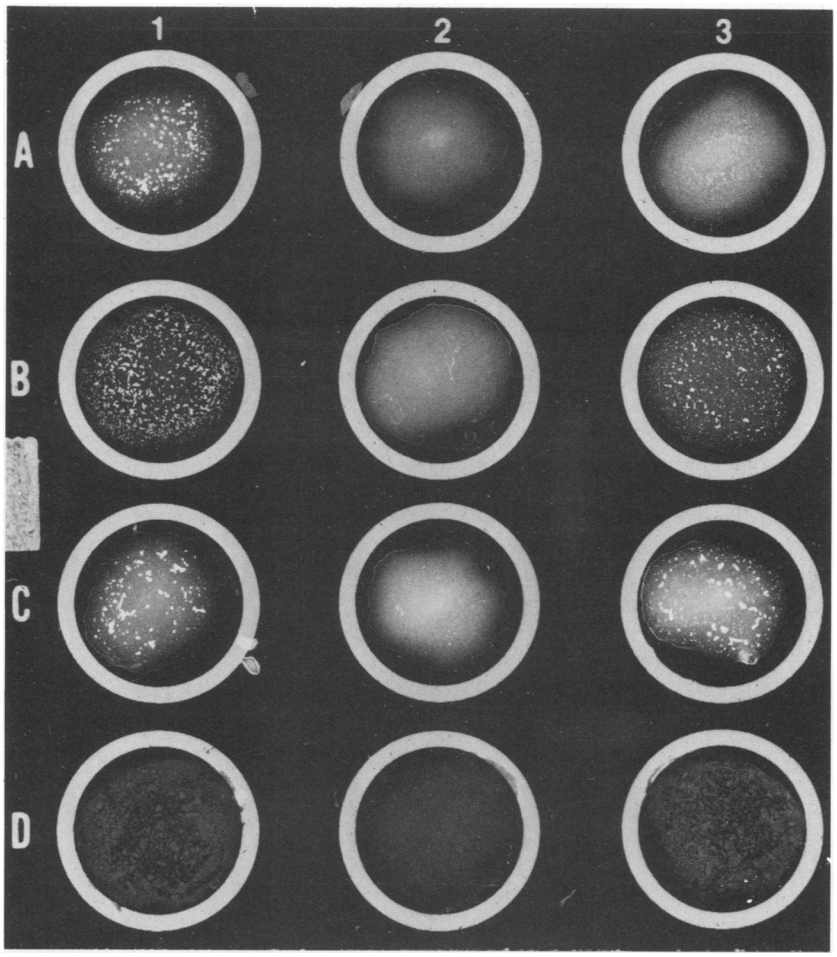

FIG. 1. Comparison of agglutination reactions with four commercial reagents for the identification of *S. aureus*. Column 1 shows reactions of *S. aureus* ATCC 25923 with each reagent, column 2 shows *S. epidermidis* ATCC 12228 reactions, and column 3 shows moderate reactions by a fresh clinical isolate of *S. aureus*. Row A, Sero STAT Staph; row B, Bacto-Staph; row C, Staphylatex; row D, Staphyloslide.

positive strains of this species have been reported (17). One of the recent clinical isolates of *S. aureus* had a negative tube coagulase test but a positive slide clumping factor reaction. This strain was negative with the Staphyloslide and Bacto-Staph reagents but positive with the Sero STAT Staph and Staphylatex reagents. *S. aureus* strains that produce clumping factor but not soluble (free) coagulase have been reported and include ATCC 25904 (2, 4). This tube coagulase-negative strain was retested at room temperature and examined at 2, 4, and 24 h but remained negative at all times (W. Landau and R. L. Kaplan, Clin. Microbiol. Newsl. **2**:15, 1980). Five of the non-*S. aureus* strains autoagglutinated in water and therefore could not be tested by the slide coagulase or the three latex tests, but four of the five could be read with the Staphyloslide test because of the negative control used with the reagent. One of the strains clumped in the negative control and could not be tested. Because it has recently been reported that some non-*S. aureus* strains autoagglutinate in water but not saline (18), we retested our strains in saline, but they also autoagglutinated in this medium.

Twenty-four of the coagulase-positive fresh clinical isolates were methicillin resistant. All 24 isolates were positive in the slide and tube coagulase tests and gave strong positive reactions with all four commercial reagents. This was in contrast to the results of Aldridge et al. (1) who reported that 12.5% of methicillin-resistant *S. aureus* yielded false-negative results in the Sero STAT Staph test. However, four of their five negative *S. aureus* isolates were positive on repeat testing. Additionally, methicillin-resistant strains which are deficient in protein A have been reported (20). Our results may reflect regional differences in strains, or the number of strains we tested may not have been sufficient to detect strains failing to react.

In conclusion, the results with all four commercial reagents closely agreed with the results of the coagulase test and with each other. These results are in agreement with recent evaluations (1, 5, 14, 16). The Bacto-Staph yellow latex and Staphyloslide erythrocyte tests facilitated visualization and interpretation. This was important with strains that did not give strong clumping. An example of a moderately clumping strain is shown in Fig. 1, column 3. The yellow Bacto-Staph latex in row B was the easiest to interpret for these strains, and the sensitized erythrocyte Staphyloslide in row D was the next easiest to interpret. The Sero STAT Staph gave the weakest reactions. Overall, the commercial reagents produced rapid results but exhibited no greater sensitivity or specificity than the tube or slide coagulase test, and the reagents are more expensive than the coagulase test. However, many laboratories have difficulty with the coagulase tests because the hydrated plasma does not maintain its activity for long unless it is kept frozen. A

TABLE 1. Comparison of coagulase tests and four commercial reagents for identification of 337 *Staphylococcus* strains—283 *S. aureus* and 54 *Staphylococcus* species other than *S. aureus*

Test	No. of isolates			Test sensitivity (%)	Test specificity (%)
	Positive	Negative	Auto-agglutinated		
Sero STAT Staph	278	54	5	98.2	100
Bacto-Staph	282	50	5	99.6	100
Staphylatex	279	53	5	98.6	100
Staphyloslide	276	60	1	97.5	100
Slide coagulase	279	53	5	98.6	100
Tube coagulase	283[a]	54[b]	0	99.6	99.6

[a] One strain with a positive tube coagulase test was negative by the slide test and each of the agglutination tests. This strain was biochemically identified as *S. intermedius*.

[b] One strain was negative by the tube coagulase, Bacto-Staph, and Staphyloslide tests but positive by the slide coagulase, Sero STAT Staph, and Staphylatex tests. This strain was biochemically identified as *S. aureus*.

reconstituted lyophilized vial must be frozen or kept refrigerated and used within 1 week of restoring. An alternative is to divide the vial contents into small samples and keep them frozen until the day of use. The time and expense of reconstituting, aliquoting, and freezing may make it more convenient to use products which are received ready for use and designed to be kept for a longer time at refrigerator temperatures before expiration. The four commercial reagents evaluated in this study produced results faster than the tube coagulase test, the reagents were more convenient to use, and the results are comparable to those of the coagulase test in sensitivity and specificity. However, but the reagents are more expensive.

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