NOTES

Multiple-Inocula (Replicator) CAMP Test for Presumptive Identification of Group B Streptococci

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Received for publication 6 September 1977

A replicator method for performing the CAMP test is described. Of 304 group B streptococci tested, 303 (99.7%) were CAMP positive. None of 1,093 non-group B streptococci was CAMP positive.

Because of their relative simplicity compared with serological identification, presumptive tests for group B streptococci are commonly used in clinical laboratories. Presumptive tests such as the CAMP test (1, 2), hippurate hydrolysis (5), pigment production (6, 11), and modifications of these (8, 10, 13) have consistently shown better than 95% correlation with group B streptococci (9). With the exception of the hippurate test, which is positive in a small percentage of group D streptococci, the presumptive tests rarely give positive results with serological groups other than group B (2, 5, 11).

The CAMP factor is a substance elaborated by group B streptococci which acts synergistically with β -hemolysin of *Staphylococcus aureus* on sheep blood agar (2). A replicator method for the CAMP test whereby large numbers of streptococci can be easily and rapidly screened and presumptively identified as group B or nongroup B streptococci was originated by one of us (R.N.J.).

Streptococci were inoculated by the replicator method as described previously (7). Briefly, streptococci to be tested were inoculated into 0.5 to 1.0 ml of Todd-Hewitt or Trypticase soy broth. After incubation for 4 to 6 h at 35°C, the inocula were placed into the wells of the seed plate of the inocula-replicating device described by Steers et al. (12). With the inoculator, the streptococci were spot inoculated onto a petri plate of 5% sheep blood agar, whereby up to 32 organisms can be inoculated upon one plate. Once the streptococci were inoculated, a β -lysine-producing *S. aureus* was streak inoculated midway between each row of spot-inoculated streptococci. The plates were then incubated aerobically at 35°C overnight. On the following morning, the plates were observed for the presence of

large, semilunar zones of hemolysis between the streptococcal and the staphylococcal streak (Fig. 1). Those streptococci producing semilunar hemolytic zones, whose length was at least three times the diameter of the streptococcal colony, were considered CAMP positive. Group A and occasionally other streptococci as well as staphylococci may produce smaller, helmet-shaped zones of hemolysis which were considered CAMP negative.

Over an 18-month period, 551 clinical isolates of streptococci were serologically grouped and subjected to the replicator CAMP test. Group A streptococci were identified by immunofluorescence (3). Groups B, C, and G were identified by counter-immunoelectrophoresis (4). In addition, 679 group D streptococci, presumptively identified by the bile-esculin test (5), were also tested by the replicator CAMP test. Also, 187 streptococci which could not be grouped by any of the above procedures were tested.

Table 1 summarizes the results of these comparisons. There were no false positive CAMP reactions, and 99.7% of the group B streptococci

TABLE 1. Number and percentage of grouped streptococci yielding a positive replicator CAMP test

Serological group	No. tested	CAMP positive	%
A	195	0	0
B	304	303	99.7
C	20	0	0
D ^a	659	0	0
G	32	0	0
No A, B, C, D, or G	187	0	0

^a Group D streptococci were presumptively identified by biochemical tests, including the bile-esculin test, but not by serological tests.

FIG. 1. Replicator CAMP plate depicting characteristic hemolytic zones of group B streptococci. A, B, D, and G refer to examples of groups A, B, D, and G streptococci, respectively. S refers to staphylococci.

gave a positive reaction. Of the 304 group B streptococci, 14 were nonhemolytic, yet all gave a positive CAMP reaction.

The replicator CAMP test is a simple, reliable, and useful method for the presumptive identification of group B streptococci.

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