NOTES

Influence of Media on Temperature-Dependent Motility Test for *Yersinia enterocolitica*

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Received 26 March 1981/Accepted 4 May 1981

The influence of six widely used media on the motility of 100 isolates of *Yersinia enterocolitica* grown at 25 and 37°C was investigated. Seven isolates were motile at 25 and 37°C in all media, and the presence of flagellated cells was demonstrated by a flagellum stain. Six isolates were nonmotile at both temperatures in all media, and no flagella were observed. Identification schemes for *Y. enterocolitica* should reflect these possibilities.

The role of *Yersinia enterocolitica* as an incitant of enterocolitis, acute mesenteric lymphadenitis, terminal ileitis, typhoid-like septicemia, erythema nodosum, and nonsuppurative arthritis has been well documented (reviewed in reference 4). As a result, procedures for the isolation and characterization of this organism are performed routinely or upon request in many clinical microbiology laboratories (1). An important diagnostic characteristic of this organism is its temperature-dependent motility. *Y. enterocolitica* possesses peritrichous flagella and, with few exceptions, is reported to be motile at 22 to 25°C, but is nonmotile at 35 to 37°C (3, 6, 9). However, isolates which are motile at 35 to 37°C have been reported (2, 5, 7), as have isolates which are nonmotile at 25°C (4, 6). Interpretation of these reports is hampered because different motility media were used. For this reason and because of the importance of the temperature-dependent motility test for the identification of *Y. enterocolitica*, we investigated the influence of six widely used media on the motility of 100 isolates of *Y. enterocolitica*.

The isolates were obtained from the Centers for Disease Control and various laboratories representing diverse geographic areas of the United States and Canada. The identity of isolates was confirmed by using established procedures (6, 9). For motility determinations, all isolates were initially streaked on Trypticase soy agar with 5% sheep blood (BBL Microbiology Systems, Cockeysville, Md.) and incubated at 37°C until isolated colonies appeared (usually 24 to 48 h). Individual colonies of each isolate were then

subcultured onto two plates containing Trypticase soy agar with 5% sheep blood. One plate was incubated at 25°C; the other was incubated at 37°C. Growth from these plates, incubated at each temperature, was used as inocula for the following motility media prepared according to the manufacturers' directions: motility GI medium, Difco Laboratories, Detroit, Mich.; motility test medium, BBL; motility medium S, Difco; motility sulfide medium, Difco; motility indole ornithine medium, BBL; and SIM medium, BBL. After inoculation, no culture was at room temperature for more than 5 min. All tests were run in duplicate, and results were recorded after 24 and 48 h of incubation. Incubation temperatures were monitored continuously throughout the study, and the incubator doors were opened only when it was necessary to record results. An isolate was considered motile when diffuse growth from the line of inoculation was observed. Flagellum stains (8) were performed for all isolates which appeared motile at 37°C and for all isolates which were nonmotile at 25 and 37°C.

Seven isolates were motile at 25 and 37°C in all media, and all demonstrated flagella when grown at 37°C. Four of these isolates demonstrated motility after 24 h of incubation; three demonstrated motility after 48 h of incubation. Conversely, six strains were nonmotile at both temperatures in all media, and no flagella were observed. The remaining 87 strains were motile at 25°C in all media tested and were predominantly nonmotile at 37°C. Eighteen isolates were motile at 25°C, but gave ambiguous or weak

reactions at 37°C in motility sulfide medium. Flagella, however, were not detected at the higher temperature. A similar phenomenon, not attributable to the same strains, was observed with motility medium S (three strains), SIM medium (three strains), and motility test medium (two strains).

The results of this study confirm other reports which have indicated that some isolates of *Y. enterocolitica* demonstrate motility at 35 to 37°C (2, 5, 7), whereas other isolates are non-motile at 25°C (4, 6). Therefore, identification schemes for *Y. enterocolitica* should reflect this possibility. Because so many ambiguous motility test results were observed with motility sulfide medium, it is recommended that this medium should not be used for the assessment of motility for *Y. enterocolitica*. Additionally, weak or ambiguous results should be confirmed with a flagellum stain or by direct wet microscopy.

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