Microbiology Test Reliability in Differentiation of Neisseria meningitidis and Neisseria polysaccharea

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We read with interest the report by Cunningham and colleagues (1) regarding the inability of matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) to differentiate Neisseria meningitidis from Neisseria polysaccharea. As highlighted in the article, these two neisseriae are also difficult to distinguish by biochemical methods. Key reactions that might be used include assessing prolyl aminopeptidase, the ability to use sucrose to produce amylopectin, and the presence of γ-glutamyl aminopeptidase (GGT); however, neither of these tests is 100% sensitive or specific (2, 3). The importance of the similarity between these two organisms was recently reinforced at our institution, when a Neisseria sp. strain isolated from the blood and cerebrospinal fluid (CSF) of a 54-year-old woman who presented with meningitis symptoms was referred to our laboratory for susceptibility testing from an outside hospital (OSH). The blood isolate was identified by the OSH laboratory as N. polysaccharea by the API Rapid NH kit (bioMérieux, Durham, NC); this result was communicated to physicians at that institution. In our laboratory, the RapidID NH panel (Remel, Fremont, CA) identified the isolate as a Neisseria sp. strain, with 57% confidence. The GGT criterion was negative by this method. The Neisseria Bacti-card (Remel) was negative for all enzymatic activity and failed to yield an identification. A latex agglutination antigen test (Remel) on the bacterial colonies yielded reactivity with the A/C/Y/W135 group but not the control or the B serogroup. Final identification of N. meningitidis was made by 16S rRNA gene sequencing (MicroSeq; ABI). The isolate was forwarded to the Los Angeles County Public Health Laboratory, which confirmed the identification of Neisseria meningitidis serogroup Y. Of importance, this isolate was confirmed to be deficient in GGT, the primary target used by most commercial kits for the identification of N. meningitidis. The isolate was also negative for polysaccharide from sucrose using polysaccharide medium and Lugol’s iodine. No special precautions were taken while working with this isolate at the OSH, after the erroneous identification as N. polysaccharea. One OSH laboratory worker had potential high-risk exposure to this isolate and was offered prophylaxis.

Because N. meningitidis is an important human pathogen spread by the droplet route, it is imperative that laboratories accurately identify these bacteria to the species level. N. meningitidis is a particular threat to laboratory workers, with an estimated incidence of meningococcal disease among microbiologists of 65 times that of the general population and an associated case fatality rate of 50% (4). This underscores the necessity of proper biosafety precautions when working with isolates that are suspicious for N. meningitidis. Current guidelines recommend that laboratorians are immunized and perform all procedures within a class II biosafety cabinet (BSC-II) for isolates from unknown sources or sterile sites (5). In our laboratories, use of ethanol or formic acid-based direct on-plate extraction, which is performed in the BSC-II, is used for all isolates suspicious for N. meningitidis or other high-risk aerosol agents when identifying organisms by MALDI-TOF (5). The unreliability of currently available diagnostic tools in differentiation of pathogenic from nonpathogenic Neisseria spp. underscores the need for laboratorians to evaluate the validity of identifications based on multiple parameters, to consider specimen type and clinical symptoms of the patient, and to practice caution when dealing with all Neisseria isolates, including when testing these by MALDI-TOF.

REFERENCES
5. Centers for Disease Control and Prevention. 2009. Biosafety in microbiological and biomedical laboratories, 5th ed. Centers for Disease Control and Prevention, Atlanta, GA.