



M032-4 Brain abscess: *Streptococcus milleri* group (*Streptococcus constellatus*) & *Peptostreptococcus magnus* (*Finegoldia magna*—Nomenclature change)

KEY: CHALLENGE SENT TO CATEGORY A LABORATORIES TO ISOLATE AND IDENTIFY AN AEROBIC ORGANISM (*STREPTOCOCCUS MILLERI* GROUP [*S. CONSTELLATUS*]) AND AN ANAEROBIC ORGANISM (*PEPTOSTREPTOCOCCUS MAGNUS*/*FINEGOLDIA MAGNA*) FROM A BRAIN ABSCESS PER THEIR LABORATORY PROTOCOL.

HISTORY This simulated brain abscess sample was sent to category A laboratories requesting set up and reporting as per laboratory protocol. CMPT recognizes that a swab of a brain abscess is not routinely accepted, however this mode of transport is currently used for proficiency samples only. Category A laboratories were expected to isolate and identify an aerobic and anaerobic organism from this challenge.

CMPT QA Internal quality testing on sheep blood agar plates incubated aerobically yielded 4+ beta-hemolytic pure growth of *Streptococcus constellatus* viable for 8 days. Anaerobically incubated sheep blood agar plates yielded 3+ tiny translucent colonies of *Peptostreptococcus magnus* viable for 8 days.

GRADING (maximum grade = 4 for reporting both organisms) As 85% of the reference laboratories reported both an aerobic and anaerobic organism this challenge was suitable to grade. All 13 reference laboratories isolated and identified the aerobic isolate as either *S. anginosus* (5), or *S. constellatus* (4), or '*S. milleri* group' (4). Eleven (85%) out of 13 reference laboratories isolated and identified the anaerobic isolate as either *Peptostreptococcus magnus* (4), or *Peptostreptococcus* species (6), or anaerobic gram-positive cocci (1).

Table 1 outlines the grades that were assigned. A grade of 4 was given to laboratories that isolated and identified both organisms; while a grade of 3 was given if the aero-

Grading—Maximum grade = 4

Identification: 62% (44/71) of category A laboratories that processed the sample received a grade of 4/4 or 3/4 for the correct identification for reporting both an aerobic and an anaerobic organism.

Notes

- ◆ Brain abscess specimens should always be cultured both aerobically and anaerobically so that all of the types of bacteria causing the infection are recovered.
- ◆ It may be clinically and epidemiologically important to accurately identify '*S. milleri*' group isolates from invasive infections since specific species have been shown to have a propensity to cause specific types of infection.
- ◆ Due to the mixed bacterial nature of brain abscess infections, two or more antibiotic agents must be used in combination for a minimum of 8-12 weeks in order to not have a relapse. Only report antibiotics that penetrate across the blood-brain barrier and that will be effective within the central nervous system.

bic *Streptococcus* species was reported along with anaerobic gram-positive cocci, refer. Laboratories that only isolated one of the organisms were given a grade of 0. Laboratories that do not process this type of specimen were ungraded.

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Aerobic Component	Anaerobic Component	Total	Grade
<i>S. constellatus</i> (14) or <i>S. milleri</i> group (10) or Group F Streptococcus (3) or <i>S. anginosus</i> (6) or <i>S. milleri/anginosus</i> group (1) - all with or without referral	<i>Finegoldia magna</i> (2) or <i>Peptostreptococcus magnus</i> (13) or <i>Peptostreptococcus</i> sp. (17) or <i>Peptostreptococcus micros</i> (2) - all with or without referral	34/71 (48%)	4
Group F Streptococcus (5) or <i>S. constellatus</i> (1) or <i>S. milleri/anginosus</i> group (1) or <i>S. anginosus</i> grp. (2) or viridans strep grp, refer (1) - all with or without referral	Anaerobic gram-positive cocci, refer (9) or probable <i>Peptostreptococcus</i> sp., refer (1)	10/71 (14%)	3
Group F Streptococcus	Anaerobic culture referred	1	1
<i>S. milleri</i> (8); <i>S. constellatus</i> (4); <i>S. anginosus</i> (6); Group F Streptococcus (5); viridans strep group (2) - all with or without referral	Anaerobes not isolated	25	0
Aerobes not isolated	<i>P. magnus</i> , <i>Peptostreptococcus</i> sp. or anaerobic gram-positive cocci—all with or without referral	1	0
Does not process this type of specimen	Does not process this type of specimen	3	Ungraded
	Total	74	

IDENTIFICATION Aerobic Component *S. constellatus* is one of three species within the '*S. milleri*' group or viridans streptococci. Based on genetic relatedness studies, the '*S. milleri*' group is currently divided into three separate species including *S. constellatus*, *S. anginosus* and *S. intermedius*¹⁻³. Isolates within the '*S. milleri*' group may be alpha-, beta- or nonhemolytic with various (A, C, F, G) or no demonstrable Lancefield antigen. Because phenotypic identification methods may not allow accurate speciation of these organisms, most laboratories simply report the presence of '*S. milleri*' group, rather than an individual species. '*S. milleri*' group isolates can be distinguished from other types of viridans streptococci by a positive VP, arginine and esculin and negative sorbitol and urease tests. In addition, mannitol fermentation results are variable³. Whiley et al.² have also identified phenotypic traits for distinguishing the three species within the '*S. milleri*' group, but no reliable test is commercially available that incorporates these enzymes (i.e. b-D fucosidase, β-N-acetylglucosaminidase, β-N-acetylgalactosaminidase, sialidase, β-galactosidase, β-glucosidase and hyaluronidase.) Recent studies have shown that DNA sequencing is the most definite method for accurately speciating the '*S. milleri*' group organisms⁴. Furthermore, it may be clinically and epidemiologically important to accurately identify '*S. milleri*' group isolates from invasive infections since specific species have been shown to have a propensity to cause specific types of infection. For this reason, it is important to refer '*S. milleri*' group isolates from invasive infections like this case to a reference laboratory for speciation.

IDENTIFICATION Anaerobic Component *Peptostreptococcus* are part of the group of anaerobic gram-positive cocci that can cause a variety of infection, usually as part of a mixture with other organisms. Specimens that require anaerobic culture should be transported in an anaerobic environment in order to reliably recover all of the types of anaerobes that may be present. Specimens should also be inoculated onto a variety of anaerobic media as soon as possible in the laboratory. Although *Peptostreptococcus* species typically stain as gram-positive cocci, sometimes they may appear elongated and resemble gram-positive coccobacilli. In addition, the Gram reaction may be variable particularly if working with a specimen that is delayed in transit or an older culture. It is therefore important to compare Gram stains from the direct specimen with a variety of media, both broth and solid and subcultures in order to accurately assess the correct cellular morphology. Vancomycin susceptibility (5-ug-disk on brucella blood agar) is also useful for establishing that an organism is gram-positive in cases where the Gram reaction is variable. Anaerobic gram-positive cocci can only be presumptively identified to the genus and species level using currently available phenotypic methods. Most laboratories performing anaerobic identification rely on a combination of antibiotic disks and commercial panels such as the RAPID-ANA or api-AN-Ident systems⁵⁻⁷. However, these

systems are highly variable in their ability to accurately identify *Peptostreptococcus* spp. although both have been reported to have a high sensitivity in identifying *P. magnus*. Laboratories must therefore correlate clinical and laboratory phenotypic test results before reporting any commercial system identification result.

ANTIBIOTIC SUSCEPTIBILITY TESTING All 13 reference laboratories reported penicillin as sensitive for the aerobic component of this challenge. Only 20 participants reported penicillin results for the anaerobic component; 14 reported penicillin as sensitive, six commented that penicillin results are 'usually sensitive'.

Aerobic Component Most laboratories that identified the '*S. milleri*' group isolate reported the organism as being sensitive to penicillin. Clindamycin and vancomycin were also commonly tested and both drugs were sensitive. However, it is important to not report drugs such as clindamycin in this case since this antibiotic does not penetrate across the blood-brain barrier. If the patient does not have a penicillin-allergy, then intravenous high dose penicillin or its equivalent (i.e. 3rd generation cephalosporin) would be used to treat a patient with brain abscess due to this organism. Alternatively, vancomycin would be used in individuals with a Type-1 allergy to penicillin. It is important to refer this isolate for antibiotic susceptibility testing if it cannot be done in-house since increased resistance to penicillin is being found amongst the viridans streptococci group including the '*S. milleri*' group.

Anaerobic Component Less than a third of laboratories reported antibiotic susceptibility results for the *Peptostreptococcus* spp. Since this isolate was recovered from a brain abscess, it is important to perform antibiotic susceptibility testing to the primary drugs that would be used to treat this infection. Most laboratories reported penicillin and metronidazole as sensitive. Laboratories also reporting clindamycin are reminded that this drug does not cross the blood brain barrier and should not be reported in this case. In this case, metronidazole would most likely be added to a high dose penicillin agent to complete 8-12 weeks of continuous treatment. Metronidazole can be given orally since it is well absorbed when administered by this route.

CLINICAL SIGNIFICANCE Spontaneous brain abscesses occur either through bacteremia from another site of infection, particularly if there is a right to left shunt in the heart, or via direct extension from a contiguous focus of infection⁸⁻¹⁰. In adults, most bacterial brain abscess are caused by serious dental infections that through bacteremia seed the brain, or via direct extension from chronically infected sinuses. Because the source of infection often originates in the mouth or sinuses, the flora recovered from a brain abscess is most often mixed aerobes and anaerobes as in this case. Brain abscesses originating from contiguous spread from infected sinuses may also contain other respiratory flora including *Haemophilus* spp.

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Specimens submitted to the clinical microbiology laboratory from a brain abscess, should always be cultured both aerobically and anaerobically so that all of the types of bacteria causing the infection are recovered. Laboratories that failed to isolate the *Peptostreptococcus* spp. in this case should review their culture procedures for handling brain abscess specimens, and those that do not have in-house capability to handle this type of specimen should immediately refer it to a reference laboratory. If referred, the specimen should be transported to the reference laboratory as quickly as possible so that all of the organisms remain viable.

Brain abscess is a serious life-threatening infection of the parenchyma and patients often present with focal neurological findings. Depending on the size and location of a brain abscess, treatment will consist of surgical excision or aspiration in combination with aggressive medical therapy^{11,12}. Patients are followed with serial CT Scans of the brain or MRI to determine the resolution of the lesion(s)¹³. Due to the mixed bacterial nature of these infections, two or more antibiotic agents must be used in combination for a minimum of 8-12 weeks in order to not have a relapse. Only antibiotics that penetrate across the blood-brain barrier must be used to treat these infections. Clinical laboratories should therefore tailor the antibiotic susceptibility report to only report antibiotics that will be effective within the central nervous system.

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ADDENDUM

Regarding the name *Finegoldia magna*, a list of current or proposed nomenclature changes and validated revisions to the genus *Peptostreptococcus* may be found on page 858 in the *Manual of Clinical Microbiology*, 8th edition. (Reference: Moncla BJ, Hillier SL. 2003. *Peptostreptococcus*, *Propionibacterium*, *Lactobacillus*, *Actinomyces*, and other non-spore-forming anaerobic gram-positive bacteria. p. 857-879. In PR Murray, EJ Baron, JH Jorgensen, MA Tenover, RH Tenover (ed.), *Manual of Clinical Microbiology*, 8th ed. ASM Press, Washington, D.C.)

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