

## Challenge M091-5

May 2009

### CSF - *Citrobacter koseri* (formerly *diversus*)

#### HISTORY

This sample was sent to category A laboratories as a simulated cerebrospinal fluid from a neonate with sepsis.

The simulated sample contained a pure culture of *Citrobacter koseri*. Laboratories were requested to identify and perform antimicrobial susceptibility testing according to their laboratory protocol.

#### CMPT QA

The sample contained a heavy growth of *C. koseri* that was viable for 19 days.

#### SURVEY RESULTS

##### Identification results (Table 1):

The sample was processed by 96% of the participating laboratories (75/78). Of those participating laboratories 96% correctly identified *Citrobacter koseri* as the microorganism cultured. As all 15 reference labs correctly identified *C. koseri*, the sample was considered acceptable for grading.

The results are consistent with the results from PT challenges ([M031-5](#): 100%, [M94-1](#): 100%). The laboratories that correctly reported *C.koseri* received a grade of 4.

One laboratory reported *Citrobacter* species and received a grade of 3. One laboratory reported *Citrobacter koseri* and *E. coli* and received a grade of 0 as CMPT QA did not demonstrate the presence of *E.coli* in this challenge.

One laboratory reported "CSF" as the microor-

ganism isolated and was graded as zero.

Consensus was achieved amongst the reference laboratories for the identification of the microorganism and antimicrobial susceptibility testing. All 15 reference labs reported ampicillin/amoxicillin resistant, 12 labs reported 3<sup>rd</sup> generation cephalosporins susceptible and 3 labs did not report. Thirteen reference labs reported gentamicin susceptible and 2 labs did not report.

##### Susceptibility Testing results (Table 2):

All laboratories (100%) performed susceptibility testing (on at least for one antimicrobial agent).

Antimicrobial agents were divided into three groups for grading:

- a) Ampicillin/Amoxicillin
- b) Aminoglycosides
- c) 3<sup>rd</sup> generation cephalosporins (CRO/CTX) + Carbapenems (IMI / MER)

Laboratories were expected to base their susceptibility report on previous critique recommendations and on CLSI guidelines for testing and reporting antimicrobial agents in CSF infections with Enterobacteriaceae.

In critique [M031-5](#), the CMPT Committee recommended testing and reporting a series of antimicrobial agents. Reporting of the antimicrobial profile is considered more relevant than reporting the antimicrobial agents alone because treatment of CSF infections with Enterobacteriaceae usually involves a combination of antibiotics.

Also, CLSI guidelines are specific about the antimicrobial agents that should not be reported

#### Grading

CLSI recommends that *Enterobacteriaceae* should be tested against ampicillin, gentamicin and a third generation cephalosporin.<sup>8</sup>

Carbapenems may have some efficacy in the treatment of *C. koseri*. Laboratories may choose to report a carbapenem either in addition to or in place of the third generation cephalosporin.

Most laboratories reported antimicrobials according to the profile, and were given a grade of 4. Some laboratories reported 2 out of the 3 groups and received a grade of 3.

Some laboratories did report according to the profile, but also reported inappropriate antimicrobials and were given a grade of zero.

**Table -1:** Identification Results for M091-5 *Citrobacter koseri* challenge

Reported results	No of labs	%	Grade
<i>Citrobacter koseri/diversus</i> +/- snnp, refer	72	96	4
<i>Citrobacter</i> species	1	1	3
<i>Citrobacter koseri, E.coli</i>	1	1	0
CSF	1	1	0
snp +/- refer	3		ungraded
<b>Total</b>	<b>78</b>		

snp: sample not normally processed

in CSF infections.

Laboratories reported antimicrobial results in two ways: those reporting only recommended agents (groups **a**, **b** and **c** above) and those laboratories reporting the recommended agents as well as antimicrobial agents that should not have been reported.

Table 2 shows the susceptibility profiles of those laboratories reporting the recommended agents. Sixty-eight per cent of the reporting laboratories reported within the recommended susceptibility profile. The laboratories that reported the three antimicrobial groups were awarded a grade of 4, labs that reported 2 out of 3 of the antimicrobial agents were given a grade of 3.

One laboratory tested the antimicrobials but did not report the results and was given a grade of zero. By not reporting susceptibility reports, the clinician is not provided with antimicrobial information for determining treatment.

31% of the laboratories reported antimicrobial agents that are not recommended for treatment of CSF infections and were given a grade of zero. See Table 3.

Reporting an antimicrobial agent that does not have the ability to adequately pass through the blood-brain barrier and into the CSF is considered unacceptable.

## IDENTIFICATION

*Citrobacter* sp. are members of the family Enterobacteriaceae. The genus *Citrobacter* is composed of three major species: *C. amalonaticus*, *C. koseri* and *C. freundii* complex (in addition to *C. freundii*, this term includes *C. braakii*, *C. gillenbergii*, *C. murlinae*, *C. rodentium*, *C. sedlakii*, *C. werkmanii*, and *C. youngae*).<sup>1</sup>

They readily grow on media used in the routine laboratory and identify well with commercial systems. Biochemical characteristics can be found in the Manual of Clinical Microbiology, 9th ed. and previous critiques .<sup>1, 11,12</sup>

## CLINICAL RELEVANCE

Neonates are at greater risk for meningitis because of their deficiencies in humoral and cellular immunity. Neonates also have less effective phagocytic function. Neonatal meningitis is most often caused by vertical transmission during labour and delivery. The most common organisms isolated are *S. agalactiae* (group B streptococcus), *E. coli*, other gram negative bacilli and *Listeria monocytogenes*. Cases of fatal meningitis due to *Enterobacter sakazakii* have been reported as a result of contaminated baby formula.<sup>2,3</sup>

The rate of mortality from bacterial meningitis has dropped in developed countries over the last thirty years. For example, prenatal screening for *S. agalactiae* has reduced the incidence of this organism as the causative agent of meningitis.<sup>2,4</sup>

*Citrobacter* sp. are found as normal flora in the female genitourinary and gastrointestinal tract.<sup>1</sup>

Sepsis involving *Citrobacter* is often polymicrobial, with reported mortality rates as high as 48% to 50%.<sup>3</sup> In the first 2 months of life, *C. koseri* has a strong predilection for affecting the central nervous system (CNS), causing sepsis, meningitis, ventriculitis and brain abscesses.<sup>5,6</sup> Neonates with the highest onset rates of infection had a mean age of 7 days.

An exceedingly high number (>75%) of these infants develop brain abscesses, with generally poor outcomes. Once a brain abscess develops, it can persist with little response to antibiotics.

Surgery and aggressive antibiotic therapy may be indicated. Severe neurological damage such as hearing loss, seizures or mental retardation and death can result.

Transmission is most often horizontal-person to person spread which can lead to nosocomial outbreaks. Vertical transmission from mother to baby can also occur.<sup>7,8,9</sup>

## CLSI Recommendations

According to the CLSI Standards<sup>8</sup>, the suggested antibiotics (group A) that should be tested and reported routinely for Enterobacteriaceae isolates are: Ampicillin, Gentamicin/Tobramycin; Cefotaxime/Ceftriaxone (CTX/CRO are tested and reported on isolates from CSF in place of cephalothin and cefazolin).

The CLSI also warns about antimicrobial agents that should not be routinely reported for bacteria isolated from CSF: agents administered by oral route only, 1st and 2nd generation cephalosporins (except cefuroxime which is administered parenterally), clindamycin, macrolides, tetracyclines and fluoroquinolones.

**Table –2:** Recommended Susceptibility Profiles reported for M091-5 *Citrobacter koseri* challenge

Amp	Aminoglyc	CTX/Mer	No of labs	Grade
R	S	S	41	4
R		S	7	3
	S	S	3	3

**Amp:** ampicillin; **Aminoglyc:** Gentamicin, Tobramycin or Amikacin; **CTX/Mer:** labs reporting Cefotaxime, Ceftazidime, Ceftriaxone plus or Imipenem or Meropenem. **R:** resistant **S:** susceptible

**Table –3:** Laboratories reporting not recommended antimicrobials for treatment of CSF infections

Amp	Amgl	CTX/Mer	Other agents reported	No of labs	Grade
R	S	S	1 <sup>st</sup> gen cep (R)	5	0
R	S	S	SXT	3	0
R	S	S	Piper-Taz (S)	2	0
NR	S	S	SXT (S) + Cip (S)	2	0
R	S	S	SXT (S) + Piper-Taz (S)	2	0
R	NR	S	1 <sup>st</sup> gen cep (R)	1	0
R	S	S	Fox (R)	1	0
R	S	S	Rifampin (R)	1	0
R	NR	S	SXT (S) + Cip (S)	1	0
R	S	NR	1 <sup>st</sup> gen cep (R)+SXT (S)	1	0
R	S	S	Fox (S) + Tet (S)	1	0
R	S	S	Cxm (S) + Cip (S) + Pip-Taz (S)	1	0
R	S	S	1 <sup>st</sup> gen cep (S) + Cip (S) + Tet (S) + SXT (S) + Pip-Taz (S)	1	0
R	S	S	1 <sup>st</sup> gen cep (S) + Cxm (S) Cip (S) + Nor (S) + NF (S) + Tet (S) + SXT (S) + Pip-Taz (S)	1	0

**1<sup>st</sup> gen cep:** 1<sup>st</sup> generation cephalosporin **SXT:** Trimethoprim/sulfamethoxazole **Piper-Taz:** Piperacillin-Tazobactam **Cip:** ciprofloxacin **Fox:** cefoxitin **Cxm:** Cefuroxime **Tet:** tetracycline **Nor:** norfloxacin **NF:** Nitrofurantoin **NR:** no report

### ANTIMICROBIAL SUSCEPTIBILITY

*Citrobacter koseri* is intrinsically resistant to cephalothin and virtually always resistant to ampicillin. It is typically susceptible to gentamicin and other aminoglycosides

Third-generation cephalosporins are more active against *C. koseri* than first- or second-generation cephalosporins. *C. koseri* is generally more susceptible to cephalosporins than *C. freundii*.

*C. koseri* is usually susceptible to imipenem.  
1,6,7

ceptibilities and inducible enzymes, traditional treatment practices may not be effective. However, third generation cephalosporins, particularly ceftriaxone and cefotaxime, are considered to be the treatment of choice.

Occasionally, neonatal infections due to inducible beta lactamase producing strains of *C. koseri* may not respond adequately. Carbapenems have been reported to be useful in treating *C. koseri* meningitis.<sup>9,10</sup> Carbapenems are commonly chosen as there are fewer side effects (eg seizures).

The Committee recommends that all Proficiency Testing samples should be processed as routine samples even when there is a staff shortage or high workload.

### TREATMENT

Traditionally, the treatment for neonatal meningitis was a combination of ampicillin and gentamicin. These agents do achieve reasonable concentrations in the inflamed meninges of the newborn and are effective against group B streptococcus, the most common organism involved in neonatal meningitis.

In pathogens, such as *C. koseri* and other gram negative bacilli, with altered antimicrobial sus-

### REFERENCES

1. Murray, P.R., Baron, E. J., Jorgensen, J.J., Landry ML, and Pfaller, M.A., . Manual of Clinical Microbiology, 9<sup>th</sup> ed. ASM Press: Washington, DC, 2007.
2. Barnett, S.M., Krishnamoorthy KS, Grieseimer DA. Neonatal Meningitis <http://e medicine. medscape. com/ article/1176960> updated Sep 29, 2006
3. Biering, G, Karlsson S, Clark NC, Jonsdotir KE, Ludvigsson P, Steingrimsson O. Three cases of Neonatal meningitis caused by *Enterobacter sakazakii* in Pow-

- dered Milk. In Journal of Clinical Microbiology Sept 1989 p 2054-2056
4. Apostol M et al: Trends in Perinatal Group B Streptococcal Disease-United States 2000-2006. From Morbidity and Mortality Weekly Report <http://medscape.com/viewarticle/588814> Published 05/18/2009
  5. Townsend SM, Pollack HA, Gonzalez-Gomez I, Shimada H, Badger JL. Citrobacter koseri Brain Abscess in the Neonatal Rat: Survival and Replication within Human and Rat macrophages. Infection and Immunity Oct 2003, p 5871-5880 American Society for Microbiology
  6. T.I. Doran, The role of citrobacter in clinical disease of children: review. *Clin. Infect. Dis.* 28 (1999), pp. 384-394.
  7. D- Kline MW, Kaplan SL. *Citrobacter diversus* and neonatal brain abscess. *Pediatr Neurol* 1987;3:178-80.
  8. CLSI Performance Standards for Antimicrobial Susceptibility Testing; 19<sup>th</sup> Informational Supplement. 2009. M100-S19 Vol. 29 No.3
  9. Agrawal D Vertically Acquired Neonatal Citrobacter Brain Abscess-case Report and Review of the Literature
  10. Straussberg R, Harel L, Amir J 2001. Long term outcome of neonatal Citrobacter koseri (diversus) meningitis treated with imipenem/meropenem and surgical drainage. *Infection* 29:280-282
- See also:**
11. CMPT critique [M031-5](#) CSF: *Citrobacter koseri* May 2003
  12. CMPT critique [94-1](#) urine sample: *Citrobacter koseri* February, 2000
  13. CMPT critique [M074-5](#) Cerebrospinal fluid: *Haemophilus influenzae*, type B February, 2008