EDUCATIONAL COMMENTARY – BURKHOLDERIA CEPACIA COMPLEX

Learning Outcomes
Upon completion of this exercise, the participant should be able to:

- discuss the clinical significance of Burkholderia cepacia complex.
- explain how to culture and identify B. cepacia complex.
- discuss susceptibility testing of B. cepacia complex.

Formerly known as Pseudomonas cepacia, Burkholderia cepacia was first recognized in 1949 by Cornell University plant pathologist Walter Burkholder as the cause of bacterial rot in onions. Genetic studies have since shown that B. cepacia is actually a group of 9 species of bacteria that are now referred to collectively as “Burkholderia cepacia complex” (Table). Initially known as genomovars I to IX, the species that comprise B. cepacia complex have distinct genetic configurations, but they cannot be differentiated by routine biochemical tests.

Clinical Significance
Burkholderia cepacia complex has emerged as a significant pathogen in patients with cystic fibrosis (CF). It easily adheres to mucin produced in the lungs of patients with CF, and infection is extremely difficult to treat both because B. cepacia is resistant to many antibiotics and because it can rapidly develop resistance to antibiotics to which it is initially susceptible. Approximately one-third of infected CF patients develop “cepacia syndrome,” a life-threatening condition characterized by rapidly progressing pneumonia and sepsis. Infected CF patients who do not develop cepacia syndrome nevertheless often experience faster decline in lung function and shortened life expectancies compared with CF patients who are not infected. Of the 9 species that comprise B. cepacia complex, B. cenocepacia and B. multivorans are most often isolated from CF patients.

Although best known for its impact on CF patients, B. cepacia complex is an important pathogen in other patient populations as well. Persons with chronic granulomatous disease are particularly vulnerable, because their white blood cells cannot produce substances necessary to kill microbial invaders. Burkholderia cepacia complex also has emerged as a cause of nosocomial (hospital-acquired) infections, although it is not usually fatal in patients with intact immune systems. It has been implicated in respiratory infections in patients on ventilators; and it has been associated with infections caused by contaminated urinary and intravenous catheters, medications, and disinfectants. Finally, B. cepacia
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complex is an opportunistic pathogen that can occasionally infect people with weakened immune systems, and very rarely, healthy people. It has been isolated from endocarditis and wound infections, from “foot rot” in troops training in Florida swamps, and from troops serving in Vietnam.

**Isolation, Identification, and Susceptibility Testing**

No special procedures are needed to collect, transport, and process specimens for isolation of *B. cepacia* complex. A Gram stain detects *B. cepacia* complex in patient specimens. On Gram stains, the bacteria appear as medium-size, straight, gram-negative rods. However, although Gram stains can detect the bacteria, they cannot identify *B. cepacia*. Identification requires culture and further testing.

*Burkholderia cepacia* organisms grow well on standard solid media such as 5% sheep blood, chocolate, and MacConkey agars; in thioglycollate and brain-heart infusion broths; and in broth-based blood culture systems. In addition, laboratories use *Pseudomonas cepacia* (PC) agar, *Burkholderia cepacia* selective agar (BCSA), or oxidative-fermentative base-polymyxin B-bacitracin-lactose (OFPBL) agar to plate respiratory secretions from patients with CF. These selective agars are formulated to enhance the growth of *B. cepacia* and inhibit the growth of other isolates. Of the selective agars, BCSA is most sensitive and specific.

On 5% sheep blood agar, *B. cepacia* colonies appear smooth and slightly raised, and they may have a dirt-like odor and produce a nonfluorescing yellow or green pigment. On MacConkey agar, they do not ferment lactose, but after 4 to 7 days they may become dark pink or red due to oxidation of lactose. Growth on selective agars may require incubation at 35°C in ambient air for up to 72 hours.

*Burkholderia cepacia complex* should always be suspected if a respiratory specimen from a patient with cystic fibrosis yields a lactose nonfermenter that decarboxylates lysine and produces a slow, weakly positive oxidase reaction. However, these biochemical reactions will not detect all strains, because 20% of strains are lysine decarboxylase-negative, and 14% of strains are oxidase-negative. Other biochemical reactions that help identify most *B. cepacia* strains are:

- utilization of glucose, maltose, lactose, and mannitol;
- α-nitrophenyl-β-d-galactopyranoside (ONPG) positive;
- ornithine decarboxylase negative;
- failure to reduce nitrate to nitrite.

Most available commercial systems can reliably identify *B. cepacia complex*, but they cannot distinguish among the 9 species that comprise the complex. Also, commercial systems may fail to differentiate *B. cepacia complex* from *B. gladioli*, *Ralstonia*, *Cupriavidus*, and *Pandoraea* spp., which can also infect patients with cystic fibrosis. Because accurate identification is crucial to choosing an appropriate therapy,
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the species identity should be confirmed by a combination of phenotypic (conventional biochemical) and genotypic (molecular) techniques whenever a commercial system identifies \textit{B. cepacia} complex in a patient with CF. Isolates can be referred to the Cystic Fibrosis Foundation’s \textit{Burkholderia cepacia} Research Laboratory and Repository, located at the University of Michigan, Ann Arbor, MI for this purpose.

Susceptibility testing on \textit{B. cepacia} complex can be done by either disk diffusion or minimum inhibitory concentration (MIC) methods. If the disk diffusion method is used, only trimethoprim-sulfamethoxazole, ceftazidime, meropenem, and minocycline should be reported. In addition to these 4 antibiotics, chloramphenicol, levofloxacin, and ticarcillin-clavulanate can be tested by MIC methods. Antibiotics other than these should not be reported even though they may be used to treat the patient, because reliable interpretative standards do not exist.

Conclusion
The bacteria that comprise \textit{B. cepacia} complex are interesting not only for their potential to harm humans but also for their potential to benefit humans. In the years that medical scientists were learning that \textit{B. cepacia} is a potential pathogen, agricultural scientists were developing what they thought were benign, environmentally friendly uses in farming. These parallel histories of \textit{B. cepacia} complex illustrate the conflict between potential benefit and potential harm that characterizes many debates about issues affecting the environment and human health.

In striking contrast to the concern \textit{B. cepacia} complex aroused in the medical field, these bacteria attracted much interest in the agricultural industry both as an ecologically friendly way to degrade long-lasting pesticides and herbicides and as a way to increase crop yields by preventing fungal diseases. For example, strains of \textit{B. cepacia} can prevent diseases caused by \textit{Alternaria}, \textit{Aphanomyces}, and \textit{Pythium} in canola, ginseng, peas, alfalfa, snap beans, and cucumbers. A strain of \textit{B. cepacia} also was developed as a seed and root inoculant for conifers to control diseases caused by \textit{Fusarium}, \textit{Pythium}, \textit{Rhizoctonia}, \textit{Cylindrocarpum}, and \textit{Botrytis}.

However, as \textit{B. cepacia} emerged as a life-threatening infection in CF patients and an opportunistic invader in other patient populations, scientists became increasingly worried that widespread use of \textit{B. cepacia} in agriculture could threaten human health. It is currently not possible to identify with certainty which strains can be safely used in agriculture. Even if this were possible, molecular genetic evidence suggests that strains that are nonpathogenic now could evolve into human pathogens. In response to these concerns, the Environmental Protection Agency in 2004 banned \textit{B. cepacia} on all raw agricultural commodities, on plant and seedling roots, and as a seed treatment.
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**Suggested Reading**


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