

Genus: Shigella

Introduction: The genus Shigella belongs to the tribe Escherichia in the family Enterobacteriaceae. This organism is named after Kiyoshi Shiga, who first discovered it in 1898. It is the causative agent of human shigellosis, which is naturally found only in humans and apes, but not in other mammals.

Classification: There are more than 40 serotypes. The classification of shigellae relies on biochemical and antigenic characteristics (O antigens). The pathogenic species are *S sonnei*, *Shigella flexneri*, *S dysenteriae*, and *Shigella boydii*.

Important Properties: Shigellae are short gram-negative rods, non-lactose-fermenting, resistant to bile salts. All shigellae have O antigens (polysaccharide) in their cell walls, and these antigens are used to divide the genus into four groups: A, B, C, and D.

Shigella can be distinguished from salmonellae by three criteria:

- They produce no gas from the fermentation of glucose

- They do not produce H₂S
- They are nonmotile.

Virulence Factors:

1. K. capsular antigen
2. O. antigen (HL)
3. Shiga toxin: with cytotoxic and neurotoxic

Pathogenesis of Shigella:

- Shigella causes bacillary dysentery, Low infective dose < 200 bacilli (can be transmitted easily unlike salmonella (More serious and virulent than salmonella
- Incubation period = 1-3 days

- Upon ingestion, the bacteria pass through the gastrointestinal tract until they reach the small intestine. There they begin to multiply until they reach the large intestine. In the large intestine, the bacteria cause cell injury and the beginning stages of Shigellosis via two main mechanisms: direct invasion of epithelial cells in the large intestine and production of enterotoxin 1 and enterotoxin 2. High fever, chill, abdominal cramp and pain accompanied by tenesmus, bloody stool with mucus & WBC and HUS are involved.

Laboratory Diagnosis:

Specimens: include fresh stool, mucus flecks, and rectal swabs for culture. Large numbers of fecal leukocytes and some red blood cells often are seen microscopically. Culture : The materials are streaked on differential media (eg, MacConkey or EMB agar) and on selective media (Hektoen enteric agar or xylose-lysine-deoxycholate agar), which suppress other Enterobacteriaceae and gram-positive organisms. Colorless (lactose-negative) colonies are inoculated into TSI agar. Organisms that fail to produce H₂S, that produce acid but not gas in the butt and an alkaline slant in TSI agar medium.

Salmonella Shigella (SS) Agar: Shigella Clear, colorless, transparent.

XLD- Agar: *Shigella flexneri* Red Colonies

TSI-Agar: Salmonella Alkaline slant/acidic butt (K/A); - H₂S and Gas-

Pseudomonads and Acinetobacter Pseudomonads The pseudomonads are Gram-negative, motile, aerobic rods, some of which produce water-soluble pigments. The pseudomonads occur widely in soil, water, plants, and animals. *P. aeruginosa* is frequently present in small numbers in the normal intestinal flora and on the skin of humans, and is the major pathogen of the group. Other pseudomonads infrequently cause disease.

Pseudomonas aeruginosa.

It is widely distributed in nature and is commonly present in moist environments in hospitals. It can colonize normal humans; in whom it is a saprophyte. It causes disease in humans with abnormal host defenses, especially in individuals with neutropenia.

Morphology and Identification:

A. Typical Organisms *P. aeruginosa* is motile and rod shaped, measuring about $0.6 \times 2 \mu\text{m}$ (Figure 16-1). It is Gram-negative and occurs as single bacteria, in pairs, and occasionally in short chains.

B. Culture *P. aeruginosa* is an obligate aerobe that grows readily on many types of culture media, sometimes producing a sweet or grape-like or corn taco-like odor. Some strains hemolyze blood. *P. aeruginosa* forms smooth round colonies with a fluorescent greenish color. It often produces the non-fluorescent bluish pigment pyocyanin, which diffuses into the agar. Other *Pseudomonas* species do not produce pyocyanin. Many strains of *P. aeruginosa* also produce the fluorescent pigment pyoverdine, which gives a greenish color to the agar. Some strains produce the dark red pigment pyorubin or the black pigment pyomelanin.

C. Growth Characteristics *P. aeruginosa* grows well at 37–42°C; its growth at 42°C helps differentiate it from other *Pseudomonas* species that produce

fluorescent pigments. It is oxidase positive. It does not ferment carbohydrates, but many strains oxidize glucose.

Antigenic Structure and Toxins:

- Pili: Adhere to epithelial cells
 - Exopolysaccharide: Anti-phagocytic property/ inhibit pulmonary clearance.
 - Lipopolysaccharide: Endotoxic effect Enzymes
 - Elastases: Digests protein (elastin, collagen, IgG)
 - Proteases
 - Hemolysins
 - Phospholipases C (heat labile): Degrade cytoplasmic membrane components
- Exotoxin A:** Cytotoxic by blocking protein synthesis
Endotoxin: like that of other gram-negative bacteria, causes the symptoms of sepsis and septic shock.
- Pathogenesis:** *Pseudomonas aeruginosa* is primarily an opportunistic pathogen that causes infections in hospitalized patients (e.g., those extensive burns), with in whom the skin host defenses are destroyed; in those with chronic respiratory disease (e.g., cystic fibrosis), in whom the normal clearance mechanisms are impaired; in those who are immunosuppressed;
- Urinary tract infection- chronic, complicated Urinary tract infection and associated with indwelling catheter.
 - Wound infection of burn sites, pressure sores and ulcers.
 - Septicaemia- “Ecthyma gangrenosum” skin lesion (haemorrhagic skin necrosis)
 - Otitis externa- Malignant external ear infection in poorly treated diabetic patients.
 - Pneumonia- Infection of the lung in patients with cystic fibrosis.

- Eye infection- Secondary to trauma or surgery.

Laboratory diagnosis: Specimen: pus, urine, sputum, blood, eye swabs, surface swabs Smear: Gram-negative rods Culture: Obligate aerobe, grows readily on all routine media over wide range of temperature (5-42 °C). Bluish-green pigmented large colonies with characteristic “fruity” odor on culture media.

In **Centrimide** agar: *Pseudomonas aeruginosa* colonies (greenish-blue in color) are medium sized and characterized by an irregular growth

In blood agar: Colonies of *Pseudomonas aeruginosa* surrounded by a wide zone of beta-hemolysis. Cultivation 48 hours in an aerobic atmosphere, 37°C.

Biochemical reactions: Oxidase positive Catalase positive Citrate positive Indole negative Produce acid from carbohydrate by oxidation, not by fermentation.

Acinetobacter *Acinetobacter* species are aerobic, Gram-negative bacteria that are widely distributed in soil and water and can occasionally be cultured from skin, mucous membranes, secretions, and the hospital environment. *A baumannii* is the species most commonly isolated. *Acinetobacter lwoffii* and other species are isolated occasionally.

A. Morphology and Identification: Acinetobacters are usually coccobacillary or coccial in appearance; they resemble neisseriae on smears, because diplococcal forms predominate in body fluids and on solid media. Rod-shaped forms also occur, and occasionally the bacteria appear to be Gram-positive.

B. Culture: *Acinetobacter* grows well on most types of media used to culture specimens from patients. *Acinetobacter* recovered from patients with meningitis, bacteremia, female genital, sputum, skin, pleural fluid, and urine, usually.

Bacillus

The *Bacillus* genus contains numerous species, many of which are not of clinical importance. Two important human pathogens – *Bacillus anthracis* and *Bacillus cereus* cause anthrax and food poisoning, respectively.

Definition

Gram-positive bacilli often arranged in chains; aerobic (some species are obligate aerobes and some facultative anaerobes); spore-forming; most species are motile; usually catalase-positive; some species are capsulate; grow over a wide temperature range on simple media.

B. anthracis

Epidemiology

Anthrax is principally a zoonotic disease and is common in some parts of the developing world.

Human infections can be classified as: non-industrial (direct human contact with infected animals) or industrial (processing of animal products by humans). Spores can survive in the soil for long periods of time and are relatively resistant to chemical disinfectants and heat.

Infection with *B. anthracis* in the UK is rare but is normally associated with handling imported animal products. Recent UK cases have occurred in intravenous drug users, probably as a result of contaminated heroin. Anthrax has been used as a biological weapon.

Morphology and identification

Can grow under anaerobic conditions; its nonmotility allows it to be distinguished from other *Bacillus* species; virulent strains are capsulate; do not produce a zone of haemolysis on blood nor a zone of precipitation on egg yolk agar (i.e. does not produce lecithinase). Identification is normally confirmed by morphological and biochemical tests.