



ESCHERICHIA COLI AND KLEBSIELLA

ESCHERICHIA COLI

21.1 INTRODUCTION

Escherichia coli (commonly abbreviated *E. coli*) is a Gram-negative, facultative anaerobic, rod-shaped bacterium that is commonly found in the lower intestine of warm-blooded organisms (endotherms). Most *E. coli* strains are harmless, but some serotypes can cause serious food poisoning in humans, and are occasionally responsible for product recalls due to food contamination. The harmless strains are part of the normal flora of the gut, and can benefit their hosts by producing vitamin K, and by preventing the establishment of pathogenic bacteria within the intestine.

E. coli and related bacteria constitute gut flora, and fecal – oral transmission is the major route through which pathogenic strains of the bacterium cause disease. Cells are able to survive outside the body for a limited amount of time, which makes them ideal indicator organisms to test environmental samples for fecal contamination.

The bacterium can be grown easily and inexpensively in a laboratory setting, and has been intensively investigated for over 60 years. *E. coli* is the most widely studied prokaryotic model organism, and an important species in the fields of biotechnology and microbiology, where it has served as the host organism for the majority of work with recombinant DNA.



OBJECTIVES

After reading this lesson, you will be able to:

- describe the morphology of Escherichia coli and Klebsiella
- describe the Cultural Characteristics of Escherichia coli and Klebsiella
- explain pathogenesis of Escherichia coli and Klebsiella
- discuss the virulence determinance

21.2 SCIENTIFIC CLASSIFICATION

Domain:	Bacteria
Kingdom:	Eubacteria
Phylum:	Proteobacteria
Class:	Gammaproteobacteria
Order:	Enterobacteriales
Family:	Enterobacteriaceae
Genus:	<i>Escherichia</i>

21.3 HABITAT

Escherichia coli are common inhabitants of the terminal small intestine and large intestine of mammals. They are often the most abundant facultative anaerobes in this environment. They can occasionally be isolated in association with the intestinal tract of nonmammalian animals and insects. The presence of *E. coli* in the environment is usually considered to reflect fecal contamination and not the ability to replicate freely outside the intestine. There is evidence however to suggest that *E. coli* may freely replicate in tropical fresh water (Bermudez and Hazen, 1988).

21.4 MORPHOLOGY

E. coli is Gram-negative, facultative anaerobic and non-sporulating. Cells are typically rod-shaped, and are about 2.0 micrometers (μm) long and 0.25-1.0 μm in diameter, with a cell volume of 0.6–0.7 μm . It can live on a wide variety of substrates.

Strains that possess flagella are motile. The flagella have a peritrichous arrangement.



21.5 CULTURAL CHARACTERISTICS

Escherichia coli or *E.coli* cells may grow on a solid or in a liquid growth medium under a laboratory condition. Solid and liquid media may have exactly the same composition except that the solid medium contains an extra 1.5% agar. Different *E.coli* clones may have different properties. Colonies growing on solid media represent different clones.



Notes

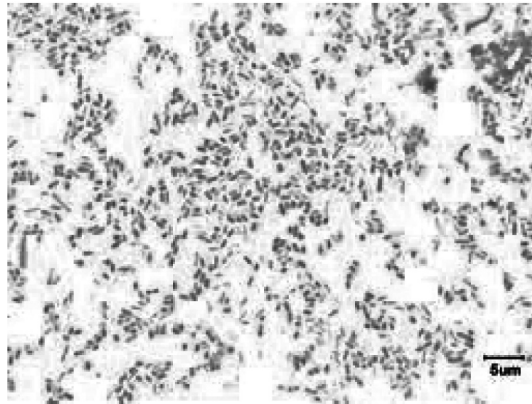


Fig. 21.1

Table 21.1

Temperature 37°C for 24 hrs	MacConkey Agar	Eosin-methylene blue Agar
Size in mm	1	1
Shape	Circular	Circular
Color	Pink	Metallic sheen
Margin	Complete	Complete
Elevation	Slightly Raised	Convex
Opacity	Opaque	Translucent
Consistency	Soft	Soft

21.6 BIOCHEMICAL REACTIONS

E. coli uses mixed-acid fermentation in anaerobic conditions, producing lactate, succinate, ethanol, acetate and carbon dioxide. Since many pathways in mixed-acid fermentation produce hydrogen gas, these pathways require the levels of hydrogen to be low, as is the case when *E. coli* lives together with hydrogen-consuming organisms, such as methanogens or sulphate-reducing bacteria.

Table 21.2

Test	Reactions
Oxidase	–
Urease	–
TSI	Acid butt, with gas, acid slant
MR	+
VP	–
Nitrate	+
Citrate	–
Indole (TW)	+
Gelatin	–

Key: + = reaction positive
– = reaction negative

Optimal growth of *E. coli* occurs at 37°C (98.6°F) but some laboratory strains can multiply at temperatures of up to 49°C (120°F).

21.7 THERAPEUTIC USE OF NONPATHOGENIC *E. COLI*

Nonpathogenic *Escherichia coli* strain Nissle 1917 also known as Mutaflor and *Escherichia coli* O83:K24:H31 (known as Colinfant) are used as probiotic agents in medicine, mainly for the treatment of various gastroenterological diseases, including inflammatory bowel disease.

21.8 ANTIGENIC AND TOXINS

The worst type of *E. coli*, known as *E. coli* O157:H7, causes bloody diarrhea and can sometimes cause kidney failure and even death. *E. coli* O157:H7 makes a toxin called Shiga toxin and is known as a Shiga toxin-producing *E. coli* (STEC). There are many other types of STEC, and some can make you just as sick as *E. coli* O157:H7.

One severe complication associated with *E. coli* infection is hemolytic uremic syndrome (HUS). The infection produces toxic substances that destroy red blood cells, causing kidney injury. HUS can require intensive care, kidney dialysis, and transfusions.



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21.9 ROLE IN DISEASES

The commonest infection caused by *E. coli* is infection of the urinary tract, the organism normally spreading from the gut to the urinary tract. *E. coli* is also the commonest cause of cystitis (infection of the bladder), and in a minority of patients the infection may spread up the urinary tract to the kidneys, causing pyelonephritis. Otherwise healthy patients in the community may develop cystitis, and patients in hospital who have catheters, or tubes, placed in the urethra and bladder are also at risk. *E. coli* is also present in the bacteria that cause intra-abdominal infections following leakage from the gut into the abdomen, as for example with a ruptured appendix or following traumatic injury to the abdomen.

E. coli bacteria may also cause infections in the intestine. Diarrhoeal infections (intestinal) are caused by a group of *E. coli* known as 'enterovirulent' (harmful to the intestines).

Overspill from the primary infection sites to the bloodstream may cause blood poisoning (*E. coli* bacteraemia). In rare instances, *E. coli* may cause meningitis in very young children.

21.10 LABORATORY DIAGNOSIS

E. coli infections can be diagnosed by the detection of *E. coli* in a laboratory test of your stool, urine, blood or other relevant sample. Infections with some types of *E. coli*, e.g. *E. coli* O157, can be detected by a serum antibody test.

Specimen used in lab for *E. coli* are

1. Urine
2. Stool

Culture

1. Isolation media- a) nutrient agar, b) MacConkey's agar c) eosin- methylene blue agar

Biochemical media

- Glucose phosphate broth
- Motility agar
- TSI slant
- Tryptone water
- Simmon's citrate agar

Escherichia Coli and Klebsiella

- Christensen's urea medium
- Nitrate broth
- Nutrient gelatin medium
- Sugars: xylose, glucose, mannitol, sucrose, maltose, etc

Reagents

1. Oxidase reagent
2. Hydrogen peroxide
3. Methyl red
4. Kovac's reagent
5. Sulfanilic acid reagent
6. O'meara reagent

Procedure

1. Perform gram staining on the given culture organisms.
2. Study motility of the organism.
3. Streak a loop-full of the organism on nutrient Agar, and MacConkey's agar
4. Keep in the incubator at 35 degree C for 24 hrs.
5. Observe for colony characters from both plates.
6. Inoculate into the various biochemical media from a single colony of the organisms.
7. Keep in the incubator at 35 degree C for 24 hrs.
8. Read the result.

Observation

1. Gram staining: gram negative bacilli
2. Motility: sluggishly motile
3. Cultural characters

Treatment of *E coli* infection

Patients, especially healthy adults, often require no treatment for *E. coli* O157:H7 since many infections are self-limited. Moreover, if required can be treated according to antibiotic sensitivity pattern.

MODULE

Microbiology



Notes



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**INTEXT QUESTIONS 21.1**

1. E.coli are Gram rods
2. Culturally E.coli are
3. E. Coli required in the media
4. E. Coli is a consistent inhabitant of the tract

21.11 KLEBSIELLA AEROGENS

Klebsiella pneumoniae can be found as a commensal in the mouth and upper respiratory tract, it is also found in moist environments, particularly in the intestinal tract of humans and animals. These are also found in plants, water and soil.

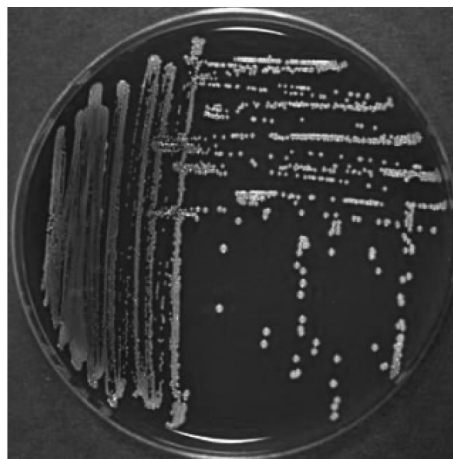


Fig. 21.2

21.12 CLINICAL SIGNIFICANCE

Klebsiella causes pneumonia, urinary infections, septicemia and other pyogenic infections. Sometimes it also causes diarrhea. *K.pneumoniae* is generally associated with lower respiratory tract infections and middle ear infections, *K.aerogenes* is generally associated with wounds and urinary tract infections.

Requirements

1. Cultural suspension
2. Isolation media - (a) nutrient agar, (b) MacConkey's agar (c) eosin-methylene blue agar

Biochemical media

- Glucose phosphate broth
- Motility agar
- TSI slant
- Tryptone water
- Simmon's citrate agar
- Christensen's urea medium
- Nitrate broth
- Nutrient gelatin medium
- Sugars:- xylose, glucose, mannitol, sucrose, maltose, etc

Reagents

1. Oxidase reagent
2. Hydrogen peroxide
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Procedure

1. Perform gram staining on the given culture organisms.
2. Study motility of the organism.
3. Streak a loop-full of the organism on nutrient Agar, and MacConkey's agar
4. Keep in the incubator at 35 degree C for 24 hrs.
5. Observe for colony characters from both plates.
6. Inoculate into the various biochemical media from a single colony of the organisms.
7. Keep in the incubator at 35 degree C for 24 hrs.
8. Read the result.

Specimen

Specimen received in the laboratory are

1. Urine
2. Sputum
3. Ear swabs
4. Wound swabs



Notes

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Escherichia Coli and Klebsiella



Notes

Observation

Organism- klebsiella aerogenes

1. Gram staining : gram negative bacilli
2. Motility : non-motile
3. Cultural characters

Table 21.3

Temperature 37°C for 24 hrs	Mac Conkey Agar	Eosin-methylene blue Agar
Size in mm	3-4	3-4
Shape	round	round
Color	Pink	pink
Margin	Complete	Complete
Elevation	Slightly Raised	slightly raised
Opacity	Translucent	Translucent
Consistency	Mucoid	Mucoid

Biochemical reactions

Table 21.4

Test	Organism K.aerogenes Reactions	Organism K.pneumoniae Reaction
Oxidase	-	-
Urease	+	+ slow
TSI	acid slant, with gas, acid butt	acid slant, with gas, acid butt
MR	-	+
VP	+	-
Citrate	+	+
Indole (TW)	-	-
Gelatin	-	-
Nitrate	+ (delayed)	+ (delayed)



INTEXT QUESTIONS 21.2

1. Enterotoxigenic E.coli (ETEC) causes diarrhea among &
2. Dysentery like diarrhea with fever are caused by
3. Haemorrhagic colitis caused by Enterohemorrhagic E.Coli may lead to
4. Klebsilla are shaped and gram bacteria
5. Pathogenesis of klebsiella is determined by
6. Klebsiella commonly causes, &



Notes



WHAT HAVE YOU LEARNT

- *Escherichia coli* is a Gram-negative, facultative anaerobic, rod-shaped bacterium that is commonly found in the lower intestine of warm-blooded organisms (endotherms).
- *E. coli* and related bacteria constitute gut flora, and fecal–oral transmission is the major route through which pathogenic strains of the bacterium cause disease.
- *E. coli* are common inhabitants of the terminal small intestine and large intestine of mammals. They are often the most abundant facultative anaerobes in this environment
- *E.coli* cells may grow on a solid or in a liquid growth medium under a laboratory condition. Solid and liquid media may have exactly the same composition except that the solid medium contains an extra 1.5% agar
- *E. coli* uses mixed-acid fermentation in anaerobic conditions, producing lactate, succinate, ethanol, acetate and carbon dioxide
- Optimal growth of *E. coli* occurs at 37°C (98.6°F) but some laboratory strains can multiply at temperatures of up to 49°C (120°F).
- The commonest infection caused by *E. coli* is infection of the urinary tract, the organism normally spreading from the gut to the urinary tract
- *E. coli* infections can be diagnosed by the detection of *E. coli* in a laboratory test of your stool, urine, blood or other relevant sample

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Notes

Escherichia Coli and Klebsiella

- Klebsiella pneumoniae can be found as a commensal in the mouth and upper respiratory tract, it is also found in moist environments, particularly in the intestinal tract of humans and animals. These are also found in plants, water and soil.
- Klebsiella causes pneumonia, urinary infections, septicemia and other pyogenic infections. Sometimes it also causes diarrhea
- Biochemically they are Oxidase negative, Urease positive



TERMINAL QUESTIONS

1. Describe the morphology & cultural characteristic E.coli.
2. Describe the virulence determinants of pathogenic E.coli
3. Describe the intestinal diseases caused by E.coli
4. List the cultural characteristics of Klebsiella



ANSWERS TO INTEXT QUESTIONS

21.1

1. Negative
2. Facultative anaerobes
3. glucose
4. intestinal

21.2

1. Infants & travellers
2. Enteroinvasive E.coli
3. Haemolytic uremic syndrome
4. Rod, negative
5. Polysaccharide capsule
6. Community acquired pneumonia, urinary tract infection & Nosocomial infection