

LECTURE: normal flora Of The GIT And Introduction To Infectious Diarrhea

[Editing File](#)

- **Important**
- Doctor's notes
- Extra explanation
- **Only F** or **only M**

"لا حول ولا قوة إلا بالله العلي العظيم" وتقال هذه الجملة إذا
داهم الإنسان أمر عظيم لا يستطيعه ، أو يصعب عليه القيام به .

OBJECTIVES:

- **Know common normal flora of the GIT**
 - **Understand the role of GIT normal flora in diseases**
 - Define and recognize the various types of acute diarrheal illness
 - Describe the epidemiology the host defenses in preventing the gastrointestinal infection
 - Explain pathogenesis by which *Escherichia coli campylobacter* and *yersinia* and their management
 - Discuss the microbiological methods used for diagnosis of each of the bacterial agents including microscopy, selective media for maximal recovery
 - Describe the pathogens, risk factors, clinical presentation and prevention of food poisoning travelers and antibiotic associated diarrhea.
 - **Name the etiological agents causing food poisoning and their clinical presentation**
-

Introduction to normal flora:

- Normal flora are microorganisms that are frequently found in various body sites in normal healthy individuals
- Constituents and number vary according to the age and physiologic status
- Able to colonize and multiply under the existing condition of different body sites.

It has some advantages such as

- Inhibit competing intruders. (تحمي الجسم من المايكرواورجانيزمز المتطفله)
- Have symbiotic relationship that benefit the host (علاقة تكافلية مع الإنسان)
- **Can cause disease in immunocompromised patients**

Normal flora - Gastrointestinal tract	Normal Flora Of GIT
<p>Ecology</p> <ul style="list-style-type: none"> ✓ Birth: sterile عند الولادة: لا يوجد نورمال فلورا ✓ Breast-fed <i>Bifidobacteria species</i> بعد ما ترضعه أمه يكتسب هذي البكتيريا ✓ Switch to cow's milk Enteric, bacteroides, enterococci, lactobacilli and clostridia ✓ Switch to solid food ✓ Microflora similar to parents يبدأ يأكل مثلهم و يحصل على نفس النورمال فلورا حققتهم ✓ Primarily anaerobic ✓ Facultative aerobes deplete oxygen ✓ Adult excretes 3×10^{13} bacteria/day 	<ul style="list-style-type: none"> • Oral cavity: contain high number of flora which vary from site to site of the mouth. • Saliva contain mixed flora :10⁸ organism /ml • Stomach : empty stomach* (sterile) has no normal flora in health due to HCL and peptic enzymes • Small intestine : very scanty except near colon • Colon of adults: 10¹⁰ org/gm stool, >90% are <i>Bacteriodes</i> (the majority are anaerobic), 10% other bacteria (which are aerobic). • Direct effect of diet composition.

-Ecology: the branch of biology dealing with the relations and interactions between organisms and their environment, including other organisms

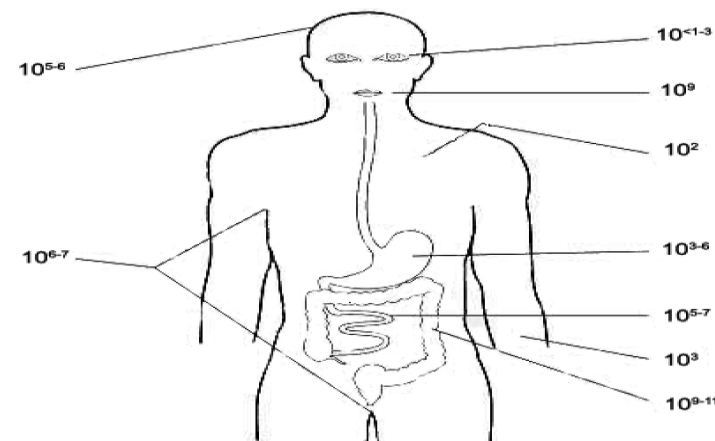
*But after eating food, it may contain some bacteria from ingested food.

This table is from 435 team

GI ecology → Varies كل لما نزلنا تحت كلما كثرت البكتيريا	
Esophagus: Microbes associated with saliva and food	Stomach: Low pH limits population numbers (10 bacteria/ml)
Small intestine Proximal small intestine (duodenum and jejunum): Sparse (< 10 ³ bacteria/ml fluid) due to acid from stomach, bile and pancreatic secretions Distal small intestine (ileum): Increases (10 ⁸ bacteria/ml) due to pH change In adult-bacteria per gram: Duodenum → 10 ³ -10 ⁶ Jejunum and ileum → 10 ⁵ - 10 ⁸	Large intestine 10 ⁹ -10 ¹¹ /ml >350 species (E.coli = 0.1% of total population) In adult-bacteria per gram: Cecum and transverse colon → 10 ⁸ -10 ¹⁰ sigmoid colon and rectum → 10 ¹¹

Location (adult)*	Bacteria/gram contents
duodenum	10 ³ -10 ⁶
jejunum and ileum	10 ⁵ -10 ⁸
cecum and transverse colon	10 ⁸ -10 ¹⁰
sigmoid colon and rectum	10 ¹¹

Increase in the number



What you need to know is that the least number is at the stomach and increases until it reaches the peak in the large intestine

*Explanation: the table includes the normal range of the normal flora population in different regions.

Normal flora (low virulence)						
Organ	Mouth	Nasopharynx	Stomach	Small intestine	Colon of adults	Colon of breast feeding infants
The Normal Flora	1-Viridans streptococci 2-Neisseria spp Moraxella, Peptostreptococcus.	Niesseria spp., Viridans sterpt. Moraxella, Peptostreptococcus.	streptococci, Peptostreptococcus, others from mouth.	scanty, variable	Bacteriodes, Fusobacterium, Bifidobacteria, Lactobacillus, enterobacteria (is Aerobic), Clostridium	Bifidobacterium, Lactobacillus (are anaerobic)

Potential pathogen (carrier)*						
Organ	Mouth	Nasopharynx	Stomach	Small intestine	Colon of adults	Colon of breast feeding infants
The Normal Flora	Candida albicans	S.pneumoniae, N.meningitidis, H.influenzae, S.pyogenes, S.aureus	none	none	B.fragilis, E.coli, Pseudomonas, Candida, Clostridium	None <small>This gives a hint about the advantages of breast feeding compare to bottle feeding</small>

*But after eating food, it may contain some bacteria from ingested food.

Role of GIT normal Flora in disease

-Many are opportunistic pathogens. **they do not cause the disease unless if there is** Example: perforation of the colon from ruptured diverticulum, feces enters into peritoneal cavity and cause peritonitis (**may lead to septicemia**) (هنا النورمال فلورا انتقلت لمكان ثاني فسببت مرض)

-Viridans streptococci of oral cavity enters the blood and colonize damaged heart valves. (**such as endocarditis**). (هذي البكتيريا مكانها الطبيعي في الفم و لكن اذا وصلت الدم بتسبب ضرر لصمامات القلب)

-Mouth flora play a role in dental caries. (ممکن تلعب دور في تسوس الأسنان)

-Compromised defense systems increase the opportunity for invasion. (أمراض نقص المناعة تعطي النورمال فلورا المجال علشان تضر الجسم) **of this normal flora to cause serious disease**

-Death after lethal dose of radiation due to massive invasion of normal flora. **فهي سلاح ذو حدين**

-E.coli: the most common Enterobacteriaceae; a facultative flora of colon followed by Klebsiella, Proteus and Enterobacter.

-Salmonella, Shigella and Yersinia are NOT normal flora of the intestinal tract.

-Some strains of E.coli, Salmonella, Shigella and Yersinia enterocolitica are able to produce diseases in the intestinal tract.

Acute diarrheal illness and food poisoning:

Introduction:

- Acute diarrheal illness is one of the most **common** problems evaluated by clinicians.
- A major cause of **morbidity and mortality** world wide.
- Most of healthy people have mild illness but other might develop serious squeals so it is important to identify those individuals who require early treatment.

Definition of diarrhea:

- Stool weight in excess of **200** gm/day or **3** or more loose or watery stools/day
- Acute < 14 days
- Alteration in normal bowel movement characterized by decreased consistency and increased frequency
- Less than 14 days in duration

Etiology:

- **Viral** the majority: **70-80%** of infectious diarrhea (**In children**) in developed countries
 - **Bacterial**: **10-20%** of infectious diarrhea but responsible for most cases of severe diarrhea
 - Protozoan: less than 10% (and may be complicated)
-

Epidemiology:

- **1.4 million deaths in study in 2010**
- 1.2-1.9 episodes per person annually in the general population
- 2.4 episodes per child >3 years old annually
- 5 episodes per year for children <3 years old and in daycare
- Seasonal peak in the winter

Risk Factors:

1. Food from restaurants.
2. Family member with Gastrointestinal symptoms.
3. Recent travel to developing countries.
4. Patient underlying illness and medication(↓Stomach acidity cyst, spores) These patient more liable to get GIT disease
5. Abnormal peristalsis.
6. Low Immunoglobulin IgA.
7. Antibiotics decrease the normal flora to less 10^{12}
8. Median infective dose (ID_{50}) so if we have low ID_{50} , the Susceptibility will be high

In countries that have disaster e.g. yemen they have more diarrheal diseases

Mostly viral , usually if it is bacteria and the patient is not hospitalized campylobacter is the organism and it is self limiting (the patients wont come to the hospital even.)

Classifications*

1. **Infectious diarrhea**: viral (Causes watery diarrhea) or Bacterial organism (Campylobacter, Shigella, Salmonella, Yersinia, vibrio Cholera and E.coli).
2. **Food poisoning**: Staphylococcus aureus (their toxin is preformed thus their poisoning effect is quick), Clostridium perfringens, Bacillus spp.(the incubation period of these 3 bacteria is Quick= 6 hours- half a day)
3. **Traveler diarrhea**: (can cause watery diarrhea) Enterotoxigenic (no invasion by the bacteria thus no pus ,and no fever because there is no inflammation) E-coli (incubation period 1-3 days and this is true for most of the diarrheal disease)
4. **Antibiotic associated diarrhea**: Clostridium difficile

Intestinal pathogens: Only in female's slides

- Invasive and Cytotoxic strains produce inflammatory diarrhea (Dysentery) with WBCs and /or blood in the stool.
- Enterotoxin –producing strains cause watery diarrhea with loss of fluid.
- Some produce not only produce local infection in GIT systemic illness due to spread to multiple organs such as enteric (typhoid) fever.

*The difference: Infectious Diarrhea : they eat infected food,

Food poisoning : They have the bacteria and then produce the toxin inside the food.

Clinical Presentation and Pathogenic Mechanism

1-Enterotoxin mediated: Here the bacteria is dead but they already released their poison

- Lack of pus in the stool (no gut invasion)
- Lack of fever (because no inflammation)
- Rapid onset preformed toxin < 12 hour
- Small intestine are affected
- Vomiting , non-bloody diarrhea, abdominal cramp
- *Vibrio cholerae, Staphylococcus aureus, Clostridium perfringens and Bacillus cereus*

All of these produce toxin which usually affect the small intestine and cause some abdominal cramp, nausea, vomiting and watery diarrhea, there is no invasion

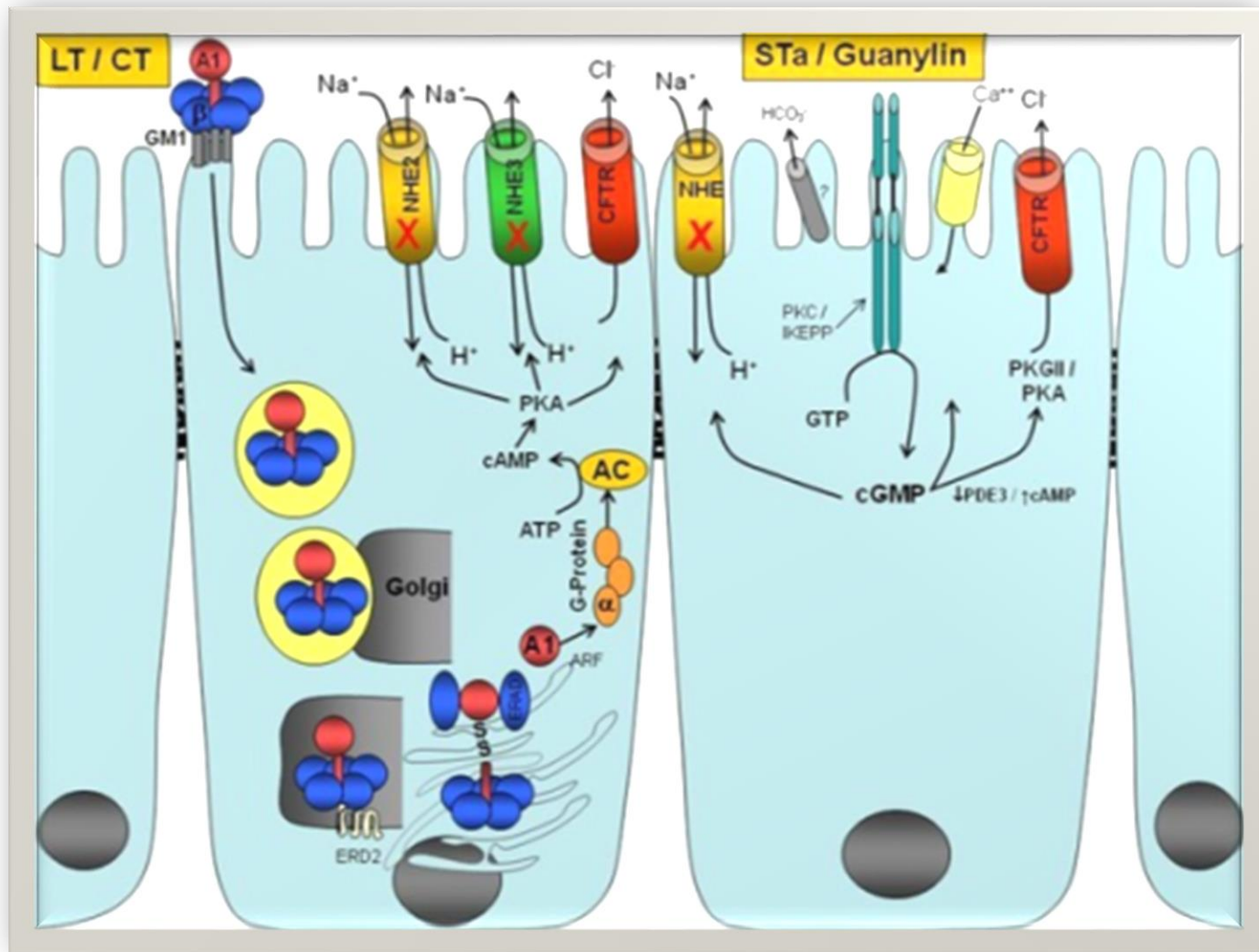
- Other viral and some parasitic infection can produce toxin but more common is via bacteria.

2-Invasive: Here the bacteria are alive and they fight and release their poisons

- Pus and blood in the stool
- Fever due to inflammation
- *Shigella, and Entamoeba histolytica (very important)*
- Affect colonic mucosal surface of the bowel
invasive mechanism → large intestine is usually affected
For enterotoxin mechanism → small intestine is usually affected
- Sometime *Salmonella spp, Campylobacter, some E-coli*
EHEC , *Yersinia and Clostridium difficile*
- Extension to lymph nodes
- Incubation period 1-3 days
- *E. histolytica* 1-3 wk
- Dysentery syndrome- gross blood and mucous with stool in small amounts after severe suprapubic pain , the pain is relieved after defecation ends. (ممكن نسألکم)
(عنها في الاختبار)
- EHEC bloody diarrhea
- If the cause is shigella we call it bacillus dysentery And if the cause is ameba histolytica we call amebic dysentery

(the parasites have longer incubation periods than others).

TOXIN TYPE	TOXIN PRODUCING BACTERIA	TOXIN NAME, IF RELEVANT
Neurotoxin Food poisoning	<i>Staph aureus</i>	Enterotoxin B
	<i>Bacillus cereus</i>	Emetic toxin
	<i>Clostridium botulinum</i>	Botulinum toxin
enterotoxin	<i>Vibrio cholerae</i>	Cholerae toxin
	<i>Enterotoxigenic E.coli</i>	Heat-labile toxin, heat-stable toxin
	<i>Clostridium perfringens</i>	Enterotoxin
cytotoxin	<i>Shigella dysenteriae type I</i>	Shiga toxin
	Enterohemorrhagic E. coli	Shiga toxins 1 and 2 Globotriaosylceramide (Gb3) 23 S r RNA in 60 Subunit
	<i>Vibrio parahaemolyticus</i>	Thermostable direct hemolysin
	<i>Campylobacter jejuni</i>	Cytolethal distending toxin
	<i>Clostridium difficile</i>	Toxin A and B
	<i>Clostridium perfringens</i>	α -toxin
	<i>E. histolytica</i>	Protozoal phospholipase A and pore-forming peptides



A1-B5 protein enterotoxin that is exported out of the bacterial cell by a type II protein secretion system

Shigella, salmonella

salmonella

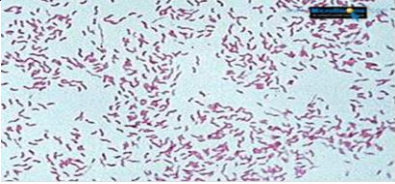
Salmonella **enterica** is the common cause of food poisoning in Saudi Arabia.

Salmonella **typhi** transmitted through human faeces

shigella

It causes local Gastrointestinal invasion and bacteremia less common in normal host.

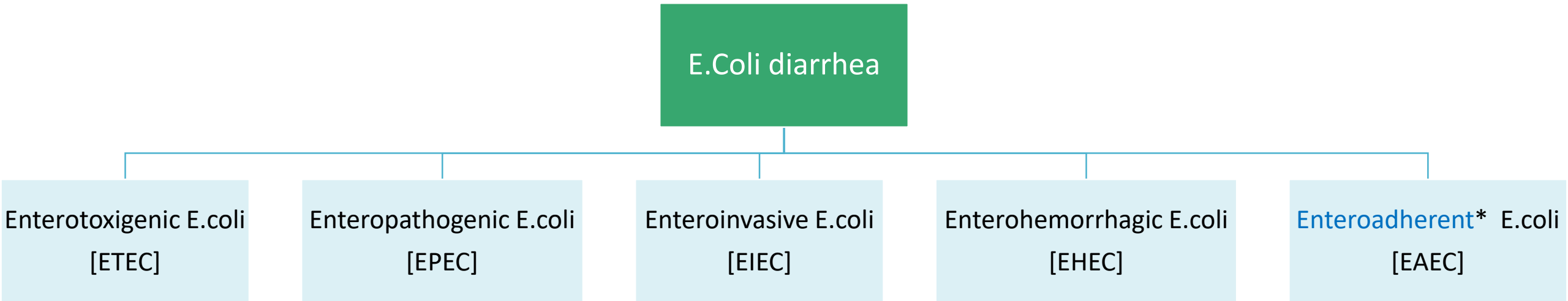
Campylobacter

	<ul style="list-style-type: none"> Gram negative curved (spiral or s-shape) bacilli . world wide infection ,more common among children Common species : <i>C.jejuni</i> (the most common), <i>C. coli</i>, <i>C fetus</i>. 	
Family	Campylobacraceae -Genus arcobacter	
Epidemiology:	Source: poultry, birds, dog , cat, →water, milk, meat, person to person can occur	
Clinically	<ul style="list-style-type: none"> IP: 2-6 days Lower abdominal pain , watery or dysenteric diarrhea with pus and blood. fever in some patients, bloody diarrhea, Nausea and vomiting are rare Self limiting 2-6 Day Chronic carrier outbreaks uncommon. <p>Complications: May lead to autoimmune disease like Guillain- Barrie' syndrome* (neurologic disease characterize by paralysis) and extra-intestinal infections eg. Reactive arthritis ,bacteremia ,lung infection and others frequently preceded by C.jejuni infection.</p>	
Laboratory diagnosis	<ul style="list-style-type: none"> Transport media Cary Blair Because it may die quickly if it does not transported to the lab Culture on CAMPY BAP media contain antibiotics Incubate in microaerophilic atmosphere (5%)O₂ ,(10%)CO₂ ,(85%)N at 42°C except <i>C.fetus</i> 37°C Identification: Gram stain/culture biochemical/Serology 	
treatment	<ul style="list-style-type: none"> Only severe cases but usually is self limiting disease and no need for treatment. Ciprofloxacin, Erythromycin or Tetracycline 	

* The history is important, if we have patient who develop sudden paralysis after he had diarrhea we can think about Campylobacter, sure after excluding of other causes such as cardiovascular problems.

E. coli

- One of the most common normal flora, and not all of them can cause diarrhea, Only about 10 -15% of strains of E. coli associated with diarrhea, Other strains associated with extra-intestinal diseases(septicaemia, meningitis in neonates & UTI).
- Based on virulence factors, clinical manifestation, epidemiology and different O (lipopolysaccharides antigen) and H (flagella antigen) serotype.
- There are five major categories of diarrheagenic E.coli:



*also called enteroaggregative e coli

	Enterotoxigenic E.coli	Enteroinvasive E.coli
The diseases	Major cause of traveler's diarrhea in infant and adult in developing countries from contaminated food and water	Produce dysentery (Penetration, invasion and distraction), Common in children
Infective doses	High : 10^6-10^{10}	10^6
Symptoms	watery diarrhea, abdominal cramps and some time vomiting	Fever, severe abdominal cramps, malaise and watery diarrhea
toxins	Has heat-labile toxin (LT) and heat-stable toxin (ST) each has two fragment (A and B). No invasion or inflammation , LT leads to accumulation of CGMP, which lead to hyper secretion of fluid with no cellular injury	-
diagnosis	No routine diagnostic method.	Diagnosis <u>Sereny*</u> test and DNA probes.
Other info	Self limiting -	-Similar to Shigella spp (Non motile, LNF) -transmission: Fecal oral route

*They take the bacteria or its toxin and they are inoculated into the eye of an animal, after a period of time , we will see that there is destruction of an animal's eye. This indicate there is EIEC

Enterohemorrhagic E.coli (very important)
The most dangerous one and most common one

The diseases	O157H7 and Non O157H7 Hemorrhagic diarrhea , colitis and hemolytic uremic syndrome (HUS)= ↓Platelet count, hemolytic anemia and kidney failure.
Prevalence	Fetal disease in young and elderly persons in nursing homes.
symptoms	Bloody diarrhea , low grade fever and stool has no leucocytes
toxins	Cytotoxin =vertoxin I and vertoxin II Similar to Stx₁ (shigotoxin I&II).
Diagnosis	Diagnosis by culture on SMAC (<i>sorbitol MacConkey agar cefixime</i>), MUG test , Vertoxin detection by immunological test or PCR.
Other info	- Undercooked hamburgers it was known as hamburger's disease, unpasteurized dairy products, apple cider, cookie dough happen to help the bacteria. -E.coli other than O157H7 can cause HUS.

	Enteroadherent E.coli Enteraggregative Ecoli	Enteropathogenic E.coli
The diseases	Pediatric Diarrheal Disease.	Causes Infantile diarrhea (bottle fed infants)
symptoms	Produce mucoid, Watery diarrhea, vomiting, dehydration and abdominal pain.	Low grade fever, malaise, vomiting and watery diarrhea
Other info	-produce Aggregative stacked bricked (like blocks), adherence to surface of intestinal epithelial cells. -resolve after 2 weeks or more	-Outbreak in hospital nurseries and day-care centers -Stool mucous but no blood. -Disrupt microvilli and intestinal absorptive function.

Yersinia enterocolitica

The diseases	Mesenteric lymphadenitis in children and septicemia in immunocompromised hosts
symptoms	Presented with enteritis, arthritis and erythema nodosum Systemic manifestation not GIT only
Prevalence	Common in Europe, USA, Canada and cat, dog, swine (chitterlings)
Characteristics	Survive cold temperatures and associated with transfusion of packed red blood cells.
Diagnosis	Growth at 25°C-30°C media <u>Cefsulodin-Igrasan-Novobiocin (CIN)</u>
Other info	Generalize infection in adult and children 1-5 yrs usually mild but in old children adult mimic appendicitis In Saudi Arabia it can be misdiagnosed with appendicitis

Clostridium difficile	
The diseases	The most common cause of Antibiotic associated diarrhea , (<i>ampicillin, cephalosporins & clindamycin</i> → is the most common one). Antibiotic use for the last 8 weeks (community acquired= outpatient) or hospital stay for at least 3 days (hospital acquired=inpatient). So the duration is very important.
Transmission	Transmit from person to person via Fecal-Oral route.
symptoms	Patient Presents with fever, leukocytosis, abdominal pain and diarrhea
toxins	Produce toxin A (enterotoxic & cytotoxic effects) and B (cytotoxic which more serious) that can bind to surface epithelial cell receptors leading to inflammation mucosal injury and diarrhea
Histological findings	Pseudomembrane With endoscopy we can see it can result (neutrophils, fibrin, and cellular debris in the colonic mucosa) and toxic megacolon.
Diagnosis*	direct toxin detection from stool by (EIA)
Treatment	Metronidazole ± oral Vancomycin and supportive treatment.
Other info	-Have been cultured from inanimate hospital surfaces. -causes Disruption of the indigenous bacterial flora of the colon

*Once it is positive, we have two situations..

- If the patient has severe symptoms and with history of antibiotic in last 3 day this indicate Clostridium difficile infection
- -if the patient is asymptomatic and does not have a history of antibiotic this indicate that it is just colonies of this bacteria without infection.

Diagnostic approach

- Duration:
 - Chronic vs acute
- Symptoms:
 - Fever, bloody, weight loss and dehydration
- Risk factors:
 - Travel ,immunocompromised , diet, medications, outbreak
 - Fever and blood do culture
 - Watery no fever symptomatic treatment
 - Hospital acquire think about *C.difficile*
- Chronic diarrhea think about:
 - Protozoa (giardia, crypto, cyclo, microsporidia, MAC)
 - Other malabsorption, lactase deficiency Or bacterial overgrowth

Lab diagnosis of diarrheal diseases due to bacterial causes

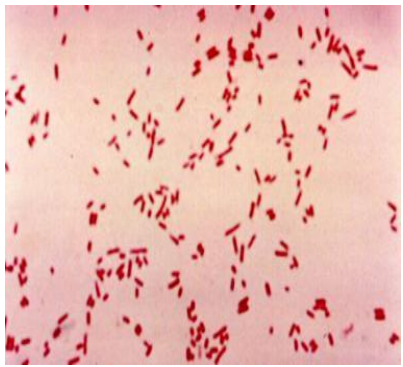
- **Stool specimen:**

Microscopy: for the presence of polymorphs or blood may help

Culture :

- ✓ on selective media for *Salmonella*, *Shigella* & *Campylobacter*. (routinely in patient with diarrhea other bacteria if the doctor request them).
- ✓ Culture for *Vibrio cholerae*, *EHEC* or *Yersinia* if suspected.

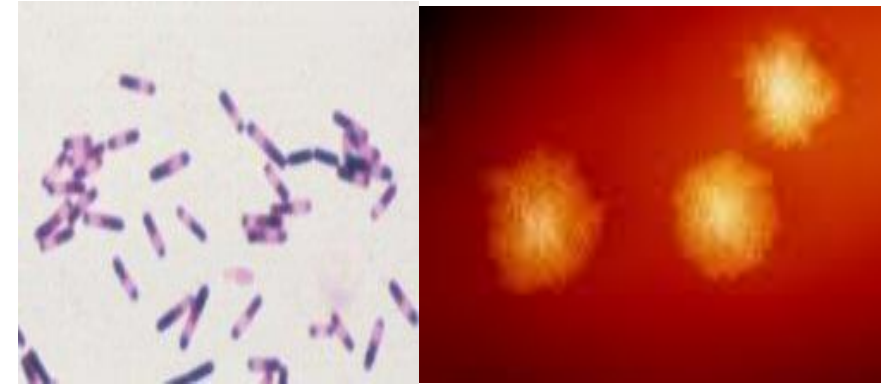
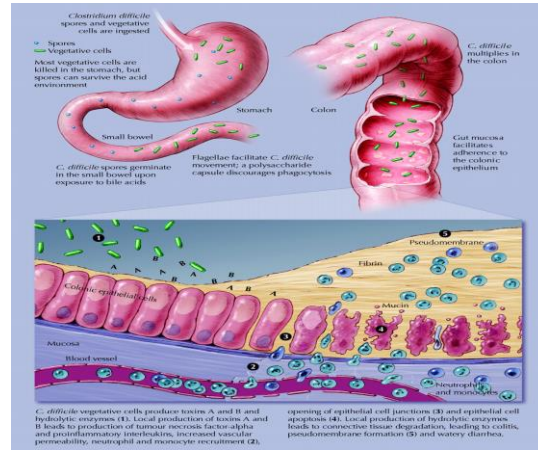
Toxin assay: if *C.difficile* toxins is suspected.



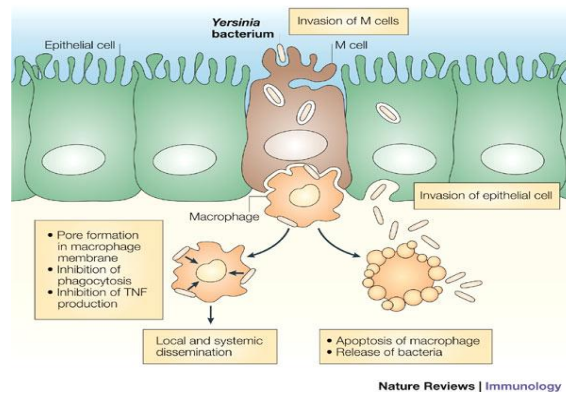
E. coli



2. Enteroinvasive
E. coli (EIEC)



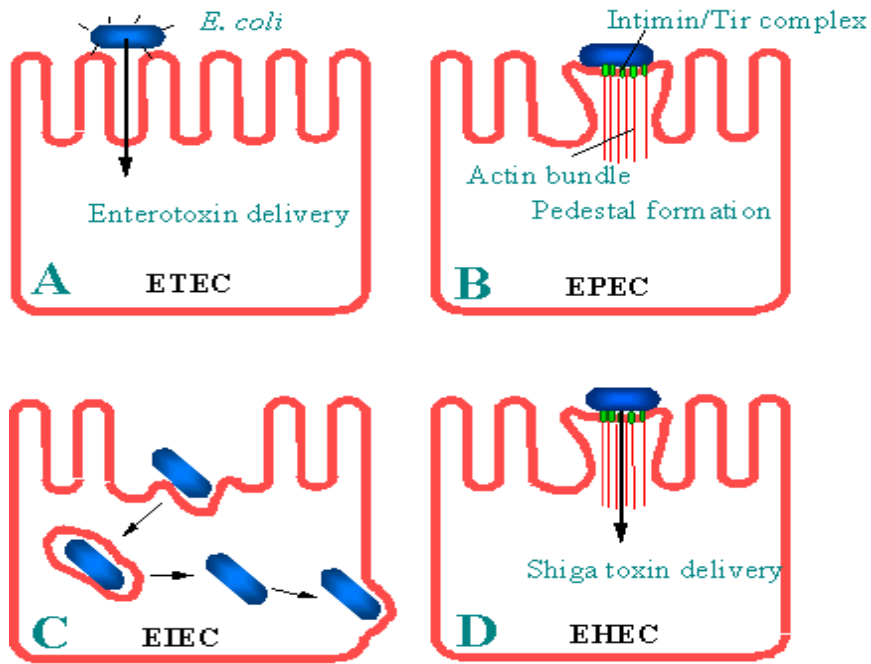
Clostridium difficile



Yersinia enterocolitica



C. difficile & pseudomembrane colitis



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Recommended from prof ali:

Type	Organism	Incubation Period	Source Risk factors	Clinical	Diagnosis	Treatment
Infectious diarrhea Invasive Dysentery Blood+mucous	Shigella gram-negative rods	1-3 Days	Contaminated Food or water with human excreta	Small amount of stool with blood and mucous and lower abdominal pain (trismus)	Culture on selective media	Ciprofloxacin
	Entamoeba histolytica	1-3 wks			Microscopy	Metronidazole
	EIEC gram-negative rods	1-3 Days			Culture and toxin detection	Ciprofloxacin

Type	Organism	Incubation Period	Source Risk factors	Clinical	Diagnosis	Treatment
Food Poisoning Watery (preformed toxin)	Staphylococcus aureus	1-6 hour	Contaminated food from human flora	Vomiting then watery diarrhea	Culture and toxin detection	Observation and supportive treatment
	Clostridium perfringens	8 to 16 hours	Food contaminated with Soil			
	bacillus cereus spore forming aerobic Gram pos bacilli	8 to 16 hours	8 to 16 hours			
Travelers	ETEC	1-3 Days	Travel	Watery diarrhea	Culture Toxin detection	ciprofloxacin
Antibiotics Associated	Clostridium Difficile	1-3 Days	Antibiotics use patient → patient	Bloody diarrhea Toxin A and B	Toxin detection PCR	Metronidazole +/- Vancomycin

Recommended from prof ali:

Type	Organism	Incubation Period	Source Risk factors	Clinical	Diagnosis	Treatment
Infectious diarrhea Non-invasive Watery +/-blood	Salmonella gram-negative rods	1-3 Days	S.typhi and S.para Human Others reptiles and snakes	Watery+/-Blood	S.typhi and S.para typhi→ typhoid Others watery diarrhea	Ampicillin or ciproflaxaci or metronidazole or trimethoprim sulfa
	campylobacter jejuni small, curved, gram-negative rods	1-3 Days	poultry	Watery+/-Blood	Special media at 42 °C at microaerophilic condition	Erythromycin
	EHEC	1-3 Days	beef	Watery then bloody diarrhea Renal failure	Culture and toxin detection	
	Vibrio cholera Comma-shape gram-negative rods	1-3 Days	Salt water	Rice water		Ciprofloxacin Doxycycline
	Yersinia Gram Neg bacilli	1-3 Days	Pork intestine	Pseudo appendicitis	Cold enrichment 25-30°C CIN (Yersinia)	Gentamycin
	Listeria Monocytogenes Gram pos bacilli	2-3 wks	unpasteurized dairy products	Meningitis in neonate and old Abortion		Ampicillin

QUIZ:

Q1-which one of the following species of E.coli causes traveler's diarrhea?

A-ETEC

B-EPEC

C-EHEC

Q2-the best drug used to treat clostridium difficile is?

A-ampicillin

B-metronidazole

C-erythromycin

Q3-A 24 year old patient presented with dysentery stool. The lab results showed gram -ve bacilli. Which one of the following is the causative organism?

A-EPEC

B-EIEC

C- EHEC

THANK YOU FOR CHECKING OUR WORK, BEST OF LUCK!



Doctors slides



Hamad Alkhudairy
Ibraheem Aldeeri
Basel almeflh



Shrooq Alsomali
Weam Babaier
Rawan Alqahtani