Microbiology lecture 3

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The last part in relation to gastrointestinal infection again we have a few types of organisms which are important as causative agent of gastrointestinal infection, these types of organisms have a special status in relation to other types which considered as classical causative agent of gastrointestinal infection, for example: Salmonella , Shigella, Enterococcus …etc.

We have here an example **Helicobacter pylori**, this is a special type of organism and the only type of organism which can reside the **stomach where it colonize in the mucus** ,and might reside in the **duodenum,** this is due to the fact that this organism produce an important enzyme called **urease ,this enzyme can manage to produce an alkaline medium surrounded by the presence of this organism due to the spreading of protein within the mucosa of the stomach and produce ammonium chloride which allaw the organism to survive.**

helicobacter pylori might colonize between 50-90% of the total human population whether they are young, adult ,men and women…etc only 2% of them develop infection from the total percentage of colonization which as you see in billions of the population of the world and might produce under certain conditions **ulceration in the lining mucosa of the stomach and produce a form of gastritis or a form of ulceration which result in duodenal ulcer and might be on the long run associated with developing of malignancy.**

So as you see this organism is important to be recognized, detected in addition to knowing the **clinical features mainly pain in the stomach ,vomiting,** a problem in the habits of eating ..etc

It might be **easily recognized** if you manage to use very few number of tests especially if you **look for the production of urease** and this can be done by direct endoscopy when you take biopsy from the infected area of stomach & duodenum ,this can be easily done in vitro by changing in the color as you have seen in urease and urea detection .

Another test, **insert** a specific device into the stomach carries **active carbon 13 or 14** which can identify the presence of urease activity in the stomach.

Culture cannot easily be done and it is not necessary , it can be done for research to look for not only the organism but also to look for the susceptibility for antibiotics.

In the last year there was a study done in **our country (JORDAN)** where they have identified 30% of the helicobacter pylori isolate **to be resistant for the metronidazole** this means that you have change the combination due to the fact that in treatment in gastritis duodenal ulcer due to this organism usually by the administration of both types of antimicrobial drugs ,one of these

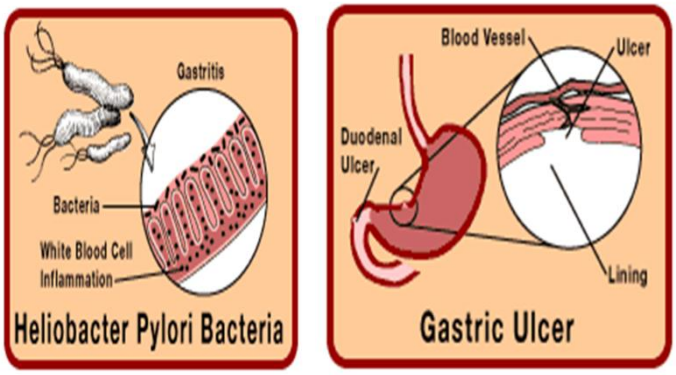
is **flagyl or metronidazole and the second can be amoxicillin or another antibiotic in addition to antiacidity suppressor inhibitor drug.**

in short, we have noticed due to wide spread of **metronidazole** the this this organism has developed **resistance.**

In our country they are using serological tests to demonstrate the presence of certain antibodies this is not helpful sometimes because it cannot demonstrate a parson who is colonized with the organism and one that has acute infection so the presence of antibodies is not enough to confirm a case especially a case of active infection , it indicates the presence of colonization therefor serological tests can be used later to control the treatment and to see if there is recovery but not for the first time to diagnose a case of helicobacter pylori infection.

in addition the source of infection is not well established but it appears that this organism is transmitted by close contact within the families by the saliva where the mother or the father transmitted the organism to their children or between parents etc.

there is no animal source for infection ,so the close contact within the family members is the only way to transfer this organism in the population.



here you can see the clinical feature of the infection and the colonization, first by **attaching** to the mucus of the mucosa and later slowly **produce cytotoxins in addition to urease which controls the stomach acidity at the site of infection where slightly begins the damage of mucosa which doesn't result in the dissemination of the organism to the blood stream ,the organism produce localized necrosis(as you see in the picture above) in the gastric mucosa and this result in the formation of gastric ulcers , there can be one(solitary or single) or more depending on the type of infection and whether it is prolonged infection.**

The gram stain can be used to demonstrate this organism if you have modify it but <u>it is better to use Giemsa stain or silver stain ,gram staining is not easy because it needs double the time order to demonstrate the presence of this spiral form bacteria.</u>

<u>Usually the flagella is present on one part, these flagella can be demonstrated by flagella stain and you might recognize only one or two or three but in fact they are 4 associated with one pole the body of the bacteria</u>

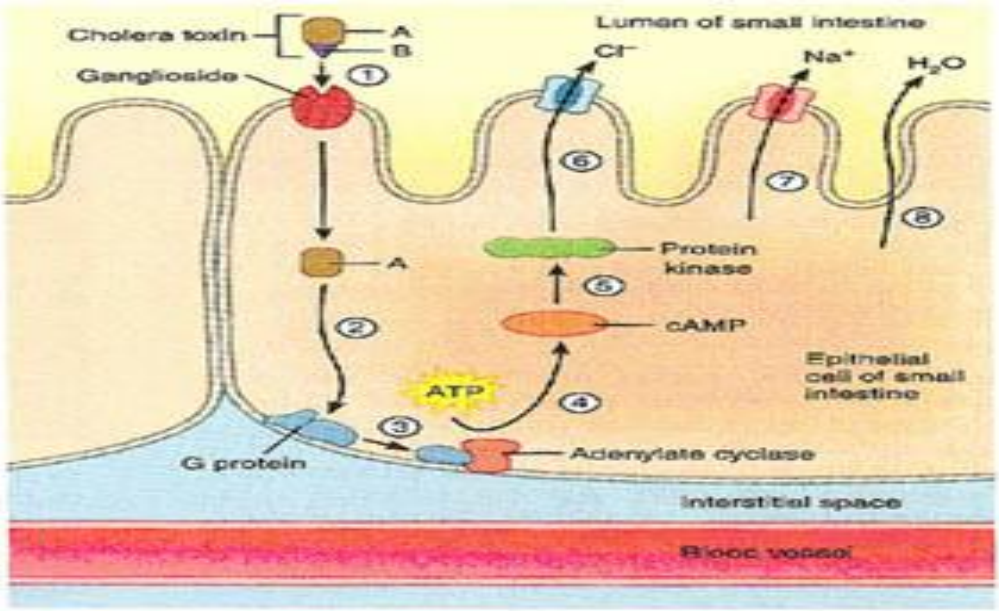
now, we move to another organism that cause diarrhea disease

<u>vibrio</u>

you have some idea about vibrio cholerae the causative agent of the classical cholera as you know already it is associated with excretion of specific type of toxin, known as cholera toxin.

Cholera toxin has been well investigated and recognized , it is composed of two specific units one called A and the second B unit , this toxin is only elaborated inside the intestinal tract(large and small intestine) during the multiplication of the organism , it is not elaborated outside the intestinal tract (NOT in vitro) !!!

The B subunit attach to specific ganglioside within the intestinal mucosa and the release of the A subunit which induces the production of cAMP which (as in E.coli) is responsible for outpouring of fluids water ,chloride,sodium ,potassium ions and this result in sever outpouring of fluids outside the body.



The toxin can cause the infected person to loss 3 liters of his body fluid in short period , which means that he will suffer from severe dehydration ,blood acidosis , shocks as kidney failures, cardiac failure and death .

Treatment :-is by replacing the lost fluids ,this can be done in association with hospitalization, giving saline as well as glucose to replace the lost fluid and you have to calculate exactly the lost and the replaced fluid to maintain normal blood pressure and body function.

During the infection with vibrio cholerae the intestine respond by production of specific Abs directed against the toxins NOT againt the cell body of the bacteria,which means that following the infection with vibrio cholerae ,the classical one 101 or the second one which is called 10329 which is mutated type of vibrio chlerae; normally ,prevent reinfection for about 1 year which means following infection with this organism there will be immunity for short period and related only for the toxin not for the cell which means this is not associated with activation of humoral Abs, humoral Abs in fact are not so important therefor the presence of vaccine (which is given orally)is related to the production of anti-toxin immunity in the small intestine and it is for short period of time.

The vaccination is recommended to be used only in endemic areas especially for travelers or for soldiers BUT not for the general population.

For general population we have to depend on prevention measures and it is well known that vibio cholerae as well as other diarrheal bacterial disease transmited by contaminated water;therefor drinking water is important to be controlled for presence of fecal material not necessary for only vibrios or salmonella or shegella to look for the presence of fecal material , which is presented usually by E.coli, if present in any tapped water or drinking water this is enough to say that it is a source of bacterial and viral infection .

The importance to v.cholerae the following :

Usually there is no sporadic cases , If you have found 1 case of v.chlerae you have to expect 100 other cases in the community which means that the outbreak of disease is related to the community and not for single person (unlike salmonellosis and typhoidal fever which are present as single cases)

If there is single cases of vibrio it is related to non v.cholerae or non 101 v.chlerae despite the fact that in biochemical test to the classical v.chlerae but differ in the agglutination with antiserum and in releasing and elaboration of cholera toxin instead to the toxin non 101 cholerae uses cytotoxin which means that it might be associated with watery diarrhea but the amount of low and not so severe and dangerus like the classical 101 vibio cholerae

So it might mimic the clinical manifiestations as the presence of watery diarrhea and there is no inflammation in the intestine but the severity of dehydration will be low incompatison with the classical one so it is easily to be recognized .

In addition to the vibrio cholerae and the non 101 vibrio cholerae which is found sometimes with the classical vibrio we have other types that are associated with GI infection and some of these might be invasive (the classical v.cholera very rarely penetrate the mucosa and reach blood vessels causing sepsis) whereas V. parahaemolyticus; it produces a form of hemolytic activity associated with specific gene responsible for invasiveness and produce cytotoxin , it also produce wound infection .

***V. parahaemolyticus* firstly it is present in water with increased amount of saline so it is a Halophilic organism.secondly, it is more found in sea water and contaminate foodbut it might if you swim in sea water and suffer from injury during swimming or some damage in the skin or the mucosa of the oral cavity the organism might produce localized infection in form of wound infection and this might later result in developing of blood sepsis.**

***V. parahaemolyticus* is associated with GI symptoms means here watery diarrhea later becomes bloody diarrhea ,vomiting and it is associated with developing of blood sepsis .this organism is rarely found in our country, found mostly in costal countries where they have more activity with sea water etc.**

It is not easy to differentiate in the lab between vibrio species without full biochemical test.

You can use specific culture medium like TCBS medium and few biochemical tests with specific antisera .

The presence of toxin is associated with the classical type 101 of v.chlerae.

Treatment:is not necessary ,the treatment is not directed towards the organism ,it is mainly rehydration and replacement of lost electrolytes

Using of antimicrobial drugs tetracyclins in adults or trimoxazole in children aims to reduce the spread of the organism in the environment and the contamination through the direct contact with water but it doesn't interfere with the course of the disease which lasted about 24 hours; during this short period intestine will produce the Antitoxin and the organism will no longer be important and even when producing toxin, the majority of toxins produced only during the overgrowth of the organism of the organism in the intestine during the first few hours then the toxin to reduce in production .

In 1976,there was an outbreak of chlerae in Jordan not necessary every one who is infected with v.cholerae to develop cholera only about 5% develop the disease , in others , their body

manage to control the infection especially if the number of the organism is not high and the body defense in addition to the normal body flora will control the increase of vibrio

Therefor the use of antibiotics at the beginning of the diarrhea will participate indirectly in the development of cholera; therefor, don't use any antibiotic if you have a mild form of diarrhea and if you have severe form of diarrhea you have to relay on the restoration of the lost fluid and this can be done by oral means at the beginning but if there is severe vomiting this cannot be done so the only way in this case is to have IV fluid .

Now, the last part of bacteria are not important in only as direct causative of GI infection but it can also produce toxins outside the body during the presence of food at room temperature or during the preparation of food whether in restaurant or in factories etc.

These types of **food born toxigenic bacteria, few numbers of them** which are widely distributed in relation to our body and in relation to the environment and easily might contaminate food during preparation and especially during the storage of food, if we have prepared any type of food containing a source of protein or sugars it might be contaminated from out hand or the environment and if we keep it without boiling, within few hours these bacteria will start to produce **extracellular toxins and the exotoxin might be associated with a form of toxication .**

- **The first type is *Staphylococcus aureus***
  It can be found in around 25%of the population carrying the *Staphylococcus aureus* in their nose and skin especially in relation to the presence of injury
  It is the first organism that reside and produce inflammatory reaction in the skin is *Staphylococcus aureus.*
  Not only staphylococcus strains produce the variation of enterotoxins , there are strains of specific bacteriophage(called lysogenic strains) which carry genes responsible for production the exotoxin.
  About 50%of the staph strains which associated with the human body with our intestine skin ..etc,it might be found in the intestine of small infants might produce these exotoxins.
  Once this heat stable exotoxins are produced in sufficient amounts (1 microgram) is enough to produce a case of intoxication
  Intoxication in relation to staph means that you have mainly vomiting after incubation period ,the vomiting is so severe associated with abdominal cramps and might be associated with dizziness and coma because it affect the centers in the brain of the patient and this feature can be for a short period less than few hours

The presence of diarrhea is not necessary and there is no fever
Mainly intoxication means mainly vomiting less diarrhea and NO FEVER.

So it is easy to differentiate between salmonella gastroenteritis and staphylococcus intoxication simply by asking the patient few questions like when was the time that you have developed the feature of vomiting?? especially if there was no fever or diarrhea then mostly it is related to staph toxins

The lab diagnosis can be done but it is not necessary, when it is done you have to ask them about the type of food which has been consumed as well as you might take a blood sample to look for the presence of toxins, but generally it is no done
Giving antibodies is not necessary because you are not dealing with any type of microorganism you are dealing with the toxin

Bacillus cereus

Aerobic spore-forming bacteria.
This organism has two types of toxins , one is Emetic Enterotoxins it is a type of exotoxin which is associated with vomiting mainly, and often this toxin is released mainly in the contaminated food and especially rice food which is not well boiled .Second,the diarrhea toxin.

The *Bacillus cereus* produce toxin type A and B ,one of them is emetic toxin and the second is responsible for diarrhea.

It is not necessary for bacillus cereus to produce both toxins at the same time, it might produce one of them only or both.

In our country 60% of bacillus cereus produce these toxins which means that they have first vomiting and then a watery diarrhea not bloody diarrhea and often the infection is mild and patients usually recover within 24 hours, therefore there is no need to give any type of antimicrobial durgs .

Clostridium perfringens

This organism has many functions and produces many toxins and enzymes responsible for a various types of diseases

***Clostridium perfringens* is an anaerobic spore forming bacteria found in the intestines of humans and animals without doing any harm under normal conditions (not as part of the flora but it can be found in the intestine for a short period of time)**

In infants and small children *Clostridium perfringens* might be considered as normal flora like clostridium difficile whereas in adults *Clostridium perfringens* if found in the intestine means you have sth wrong in the intestinal flora and especially in the use of antimicrobial drugs because there will be shift to type of aerobic and anaerobic bacteria and might *Clostridium perfringens* survive on the cost of other types .

***Clostridium perfringens* if contaminate food especially food originated from dairy products (like milk, cheese …) or in association with vegetation, It might produce outside the body again a type of toxin similar to the exotoxins of *Bacillus cereus* which means it is associated mainly with vomiting and might at the same time produce the other toxin which is responsible for diarrhea but the toxin that is responsible for diarrhea is secreted during the presence of *Clostridium perfringens* in the intestine which means if you eat a type of food with large number of *Clostridium perfringens* you might later develop diarrhea once the organism produce the diarrheal toxins in the intestines and Not outside the body .**

Outside the body you have intoxication and inside the body you have diarrhea follow the production of the toxins in the intestine.

In addition *Clostridium perfringens* might produce other types of toxins together with the enterotoxins (there is numbers for each of these but NOT NECESSARY TO BE MEMORISED) might be associated with inflammatory reaction and necrosis which means that the type of diarrhea is boody and interfere with the deferential diagnosis for other organisms like shegella or nontyphoidal salmonella which might produce this feature.

So there is vomiting, watery diarrhea , watery –bloody diarrhea in association with this organism

Produce a feature known as necrotizing enteritis and this might give the impression that the patient is suffering from pseudomembranous colitis which is caused by clostridium difficile

Necrotizing enteritis can be so severe and fatal in immunocompromised patients or eldery patients, their work should be identified, and the patient should be treated with antimicrobial drugs in order to reduce the severity of the infection while in the case of vomiting and diarrhea there is no need to give antimicrobial drug .

Clostridium botulinum

Anaerobic spore forming bacteria ,widely distributed in the intestine of large animals might be found in the intestine of human.

Generally *Clostridium botulinum* cannot produce potent toxins inside the intestinal tract , the dangerous thing about it is the toxin that it produce outside the body called the Botulinum toxin which is divided into 6 types there are strains that produce more type a than c

In general the importance of this toxin,first it is only produce by the presense of high amount of proteins under anaerobic conditions like meat fish..etc canned food and here the toxin in 2-3 NANOGRAMs are enough to produce a case of botulism which is so severe an fatal especially if the patient hasn't treat it with specific antisera to neutralize the toxin.

The fatality might reach 60% but we are lucky that in our country we rarely have a case of botulism

The mechanism of action is very interesting and important ,therefor the botox is used in cosmetic surgery and in certain types of nerve system infection.

In addition it can be used in medical treatment of other types of diseases related to the spasm of muscles ,cosmetics..etc.

The mechanism of action:-

Once this toxin has been attached to presynaptic nerve ending of the peripheral nervous system & cranial nerves system it tends to inhibit the transfer of neural transmition due to the inhibition of the release of ACH ,this mechanism resulted within few minuts in developing of spasm in many parts of the body especially in the respiratory tract ,produce a form of paralysis ,respiratory failure ,cardiac failure and death.

There is available antitoxin serum but it is not present in our country , it is present in few countries of the world , most cases has ended with the death of the patient .

Clostridium difficile

It is part of the intestinal flora in 20-30% of the population especially in hospital

If there is use any type of antibiotic especially the clindamycin and any other wide spectrum antibiotic like the amoxicillin ampicillin and second generation cephalosporins and the third generation all these antibiotics can affect large number of the intestinal flora ,decreasing it and allowing the organism to increase in number and to produce a toxin known as difficile toxin .

The difficile toxin can be type a or type b

Recently there has been newly recognized toxin known as biliary toxin the presence of any of these toxins will result in severe inflammatory reaction in the large intestine resulting in pseudomembranous colitis which means that the patient will suffer from severe bloody diarrhea ,treated by stopping the use of the offending drug and replace it by other drugs which inhibits the growth diviation toward clostridium difficile like metronidazole and vancomycin.

The end

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