Non-Bacterial Agents of Foodborne Illness

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Microbial Ecology in Health and Disease 2000; Suppl 2: 105–109

Many species of worm parasites and protozoans live in the gastrointestinal tract, but only a few are cause of serious gastrointestinal disorders. Transmission of intestinal parasites is mediated by the release of cysts, eggs or larvae, usually in stools. Some species are acquired through contaminated food or water or directly from the diseased person or animal. Gastrointestinal disorders occurring range from a mild diarrhoeal disease to an acute or chronic illness or, more rarely, to a life-threatening disease caused by spreading of parasites into the blood and organs.

Planktonic and benthic algae can produce very toxic compounds which may be transported to filter-feeding shellfish or to herbivorous fish which are food for larger carnivorous fish. Along the food chain they can be concentrated, and fish caught for human consumption can be toxic.

In addition, some moulds have the ability to produce toxic metabolites, such as mycotoxins, in foods and are associated with human disease, for example gastroenteritis or cancer.

Mushrooms also are considered as a traditional source of food but there are a few species considered to be deadly poisonous, including *Amanita phalloides* and *Cortinarius*.

PLATYHELMINTHS

Platyhelminth is derived from the Greek word meaning 'flat form'. They include 2 groups of parasites: Trematoda and Cestoda.

Trematoda

Fasciola hepatica (liver fluke) is the most common Trematoda (1). The mature stage develops in humans, sheep or cattle. It is a leaf-like animal of 25 mm, which can be found in the bile duct after feeding on the liver. It produces large eggs with an operculum, which are secreted in faeces after passing from the bile duct and the gut.

In water, the eggs produce a motile and ciliate miracidium, which infects the water snail (*Limnea tranculata*) as a secondary host. The organism is released in the larval stage (cercaria) which encyst on water cress and can survive for up to one year. Cysts will develop further if they are swallowed by animals or humans after eating raw or undercooked watercress.

The main symptoms of the disease produced include fever, tiredness, loss of appetite with pain, discomfort in the liver region of the abdomen.

Laboratory diagnosis depends largely on finding the eggs in faeces, biliary or duodenal fluids.

Cestoda

Of the human tapeworms the beef tapeworm *Taenia saginata* (2, 3), transmitted through infected beef, is the most widely distributed. Another human tapeworm, *Taenia solium*, is associated with consumption of pork meat.

Human is the definitive host. The larval forms of *T. saginata* have to develop in cattle and finally infect humans through the consumption of undercooked beef. The larval stages of *T. solium* can develop in human or pigs.

The larval stages called *cysticerci* develop in many tissues including muscle tissue. The mature form develops only in the human intestine and releases eggs, and there will be an invasion of body tissues (cysticercosis).

Diagnosis involves finding *Taenia's* segments or characteristic eggs in the stool.

Taenia echinococcus, a small tapeworm, is widely distributed and causes a disease called hydatidosis. The dog is the definitive host, rarely man. The larval forms develop in sheep, ruminants and man and infect dogs who eat contaminated meat or viscera. The mature forms develop in dog or man. The adult worm consists of a scolex (head) with a neck segmented to proglottids. *Taenia* sheds proglottids-containing eggs. When proglottids reach soil, they release their eggs containing embryos. When eggs are digested by herbivores, the embryos are released and

 ISSN 1403-4174

penetrate the gut wall, and are carried to striated muscles heart, diaphragm and organs, where they are transformed in larvae (hydatide).

Diagnosis involves finding the characteristic proglottids or eggs in the patient's faeces.

NEMATODES

Nematodes derive their name from a Greek word meaning 'roundworm' because of their shape.

Trichinella spiralis (4, 5) is a widely spread Nematode responsible for the disease trichinellosis. The parasite has no free-living stage, but it passes from host to host, including humans and pigs.

The disease is acquired from consumption of infected raw or poorly cooked pork products, especially sausages. Infection starts by the consumption of muscle tissue containing encysted larvae in a cyst with a calcified wall. In this state, the larvae can survive for many years before eaten by a second host. In the second host, the larvae are released and mature in the intestinal lumen. The female worm produces more than a thousand larvae, which penetrate the gut wall and are carried to specific muscle tissues, in which they encyst.

Few days or month after eating the infected food the symptoms of the disease appear in 2 stages: The first phase starts when larvae are invading the intestinal wall, producing abdominal pain, nausea, and diarrhoea. The second phase occurs when larvae encyst in muscle tissues, causing muscle pain and fever.

PROTOZOA

The flagellated protozoan Giardia lamblia (6, 7) causes a non invasive diarrhoea of the small intestine.

The transmission of the disease is associated usually with water or vegetables or is transmitted from person to person by poor hygiene. The organism survive in food and water as cysts. The infective dose is very low and, once ingested, the gastric juice aids the release of the flagellate active protozoa (trophozoites), which have 8 flagella and 2 nuclei. Recent data suggest that *Giardia* may also be transmitted sexually.

The symptoms of the disease involve abdominal cramps, nausea and diarrhoea. The cysts are resistant to chlorination processes used in most water treatment systems and are killed by the normal cooking procedures.

Laboratory diagnosis depends largely on the microscopic identification of cysts or trophozoites in faeces. Trophozoites are present in stools during the acute phase of the diarrhoeal disease.

Entamoeba histolytica (8) is another protozoan found world-wide, responsible for the amoebic dysentery.

The disease is transmitted by the faecal-oral route, when poor hygiene conditions prevail. A number of outbreaks from infected food are reported. The organism survives in the environment in an encysted form. A person with amoebic dysentery pass up to 50000 cysts per day.

Most infections are without symptoms (minuta form). The illness start with mucous and bloody stools, due to the ulceration of the colon, accompanied by diarrhoea, abdominal pain, fever, and vomiting (histolytic form). Hepatic and lung abscesses are caused by migration of the organism. Infections with commensal forms of the amoeba are asymptomatic.

Laboratory diagnosis depends on repeated stool examinations for the presence of the characteristic 4-nucleate cysts in faeces. Immunological tests are also available commercially.

SPOROZOID PROTOZOA

The genus Sarcocystis (9) include 2 species: *S. hominis* associated with disease in pig and *S. suihamonis*.

Sarcocystis species have two-host parasites, the definitive host (cat, dog, man) in which sexual reproduction occurs and an intermediate host (cattle, sheep, pig) in the tissues of which the asexual cysts are formed.

Mild diarrhoea and nausea are reported as symptoms of the disease. Adequately cooked beef and pork meat lose their infectivity.

Cryptosporidium

The species *C. parvum* (10–12) is of particular importance. It is found in small numbers in foods and the infective dose required to cause disease remains very low. Raw sausages seems to be the mainly incriminated food.

The whole life cycle of *Cryptosporidium* take place in a single host, which can be man or a farm animal. The ingested thick-walled cysts enter the small intestine and release sporozoites which penetrate the enterocytes where sexual reproduction leads to the presence of zygotes. About 80% of zygotes form oocysts that sporulate within host cells. The remaining oocysts are shed in faeces, and the infection is transmitted when oocysts are ingested.

Many different routes for transmission of the disease have been reported, including zoonotic, person-to-person, water, nosocomial and food transmission.

The diarrhoeal disease lasts usually 9 to 23 days. Immunocompromised persons show a more pronounced disease including profuse watery diarrhoea (30 days), fever (39°C), abdominal pain, nausea and vomiting.

Concentration techniques and modified acid-fast staining are necessary to identify the oocysts of this protozoan.

Toxoplasma gondii

The definitive host is the domestic or wild cat but other vertebrates and humans can be infected, when oocysts are shed in faeces contaminating the eating grass or foodstuffs.

Foodborne infection in man occurs by consumption of raw or undercooked meat or unwashed vegetables. The disease is initiated upon ingestion of oocysts from cat

faeces which release 8 motile sporozoites in the intestine. They multiply in tachyzoites and form tissue cysts with bradyzoites. The bradyzoites, if the cysts are broken or in immunosuppresed persons, can multiply as tachyzoites and cause an active infection. Bradyzoites are transformed into sexual cells (macro- and microgamets), which give oocysts and spread the infection (13, 14).

The disease is usually symptomless. Congenital toxoplasmosis and toxoplasmosis in immunocompromised persons (AIDS) is more severe. The usual symptoms are lymphadenopathy and mild fever. In immunocompromised persons, encephalitis is observed. Only the 10% of newborns from infected mothers during their pregnancy (especially the first trimester) develop injuries of the central nervous system.

The most common problem in the newborn is chorioconjunctivitis, hepatosplenomegalia, hydrocephaly, microcephaly.

Infections are diagnosed by demonstrating rising antibody titres in the patient's serum samples.

TOXIGENIC FUNGI

Mycotoxins of Aspergillus

Aflatoxins

Aflatoxins of *Aspergillus* are immunosuppressive by inhibiting the protein synthesis.

In 1959, a contaminant in the groundnut meal given to turkeys caused the turkey X disease. It was an aflatoxin produced by *Aspergillus flavus* growing on the groundnuts (15).

Aflatoxin is acutely toxic and carcinogenic causing serious disease states such as carcinoma in rainbow trout or hepatitis X in dogs. *A. flavus*, *A. parasiticus*, *and A. nomius* produce also aflatoxins (16). Aflatoxin contamination is an essential problem of storage of harvested products. Conditions of high humidity and warm temperature give rise to high levels of aflatoxins. Maximum tolerated levels in foods for human consumption is 30 ωg/kg as recognised by the FAO and WHO.

Aflatoxin B_1 is the most toxic aflatoxin. It is associated with liver cancer and cirrhosis of the liver. The compound is metabolised in the animal and the toxicity is influenced by its metabolic activity. The cow is able to hydroxylate the molecule and excrete aflatoxin M_1 in milk. In this way there is contamination from milk and milk products.

The production of an epoxyde could be the key to both acute and chronic toxicity. Animals producing epoxyde are responsible for the chronic toxicity and carcinogenicity and animals not producing epoxyde for the acute toxicity.

Very young children may be exposed to aflatoxins, before they are weaned because mothers had consumed aflatoxin in their food and were secreting aflatoxin M_1 in their milk.

Ochratoxins

Aspergillus ochraceus produces a nephrotoxin called ochratoxin A (17) (18). It is a contaminant of cereals and it is known to be the aetiological agent in kidney disease in pigs and it can pass through the food chain in meat products to man. Ochratoxins are immunosuppressive by inhibiting RNA synthesis.

Balkan endemic nephropathy is a serious disease occurring by eating mould-ripened hams stored for long periods of time.

Other Aspergillus toxins

Aspergillus versicolor produces sterigmatocystin in foods (Edam and Gouda cheeses when stored for long periods of time). *Aspergillus versicolor*, *Aspergillus flavus*, and *Penicillium cyclopium* (17) produce the cyclopiazonic acid responsible for the Kodua poisoning in India from consumption of kodo millet infected with *A. flavus* and *A. tamarii*. The symptoms of the disease include nervousness, depression, spasms, tremors, and giddiness.

Mycotoxins of Penicillium

Penicillium is more known as a spoilage mould (18). *P. italicum* and *P. digitatum* cause the blue-green mould of oranges, lemons; *P. expansum* causes the soft rot of apples, moulding of jams, bread and cakes and, finally, *P. roquefortii* and *P. camembertii* create the mould-ripened blue and soft cheeses.

Mycotoxin patulin

Penicillium expansum, *Aspergillus clavatus* and *Byssochlamys* (19) produce patulin, the toxicity of which is non-demonstrated. If this substance is found in a fruit or apple juice, it is an indicator of the poor quality of fruit.

P. citrinum produces citrinin which is a nephrotoxic metabolite and is responsible for the yellow rice disease in Japan. Several species of *Penicillium* produce toxic metabolites in rice. The moulded rice is yellow because of the presence of yellow toxic metabolites. *P. toxicarium* and *P. islandicum* are also involved.

Mycotoxins of Fusarium

Alimentary toxic aleukia

This disease is known as septic angina and acute myelotoxicocis (20). In Russia during the years of famine 1942–47 the consumption of cereals moulded by *F. sporotrichioides* and *F. poae* was increased and showed elevated levels of Trichotecenes (T-2 toxin), which are powerful toxins. These compounds are immunosuppressive (inhibit translation) and the patient usually dies of infectious diseases.

The symptoms of this disease involve damage of the mucosal membranes, bleeding, vomiting, diarrhoea, damage of the bone marrow and the haematopoietic system, and anemia.

Deoxynivalenol (DON) and other trichothecenes. Fusarium graminarum and *Fusarium nivale* (DON) (20) cause the red-mould disease. Symptoms include nausea, vomiting and diarrhoea, after consumption of rice, rye, wheat, barley, oat.

Zearalenone. *F. graminarum* and *F. culmorum* (20) produce an oestrogenic mycotoxin causing vulvovaginitis in pigs fed on mouldy maize.

Oesophageal cancer. The oesophageal cancer shows high incidence in Northern China and Southern Africa. *F. moniliforme* mould of cereals produce mycotoxins (20). Among these mycotoxins, moniliformin is responsible for the disease of rice, and equine leucoencephalomalacia, liver cancer in rats and the human oesophageal cancer.

Mycotoxins of other fungi

Claviceps purpurea (20, 21) is a parasite of cereals causing the ergotism disease from ergots alkaloids. Finger and toes of the patient become gangrenous and necrotic.

The sweet potato (*Ipomea*) is damaged by plant pathogens producing phytoalexins which are hepatotoxic. Ryegrass staggers is responsible for a disease in sheep. *Acremonium loliae*, which is an endophytic fungus, produce mycotoxins called lolitrems. In this last case, it is possible to eliminate the endophyte by careful heat treatment of seed but it will be susceptible to insect damage.

TOXIGENIC ALGAE

Dinoflagellate toxins

Gonyaulax catenella and *Gonyaulax tamarensis* are responsible for the paralytic shellfish poisoning, during the period when these algae form blooms (20, 21). The illness is serious with a high mortality rate. The toxic metabolites are saxitoxin and gonyautoxin, which block nerve transmission causing symptoms are tingling, giddiness, incoherent speech, and respiratory paralysis.

Ptychodiscus brevis is responsible for the neurotoxic shellfish poisoning which is a diarrhoeic disease caused by the lipophilic toxin, dinophysistoxin.

Gambierdiscus toxicus causes the cignatera poisoning including symptoms such as nausea, vomiting, diarrhoea, neurosensory disturbances, convulsions, and muscular paralysis. The disease can be life-threatening. The algae produce ciguatoxin, which is concentrated along the food chain and when the man eats fish such as moray, eel or baracuda, the disease develops.

Cyanobacterial toxins

Microcystis, *Anabaena* and *Aphanizomenon* (20, 21) can form blooms in water and cause deaths in animals who drink this water. In humans outbreaks of gastroenteritis are reported.

Microcystis aeruginosa produces toxic metabolites called cyanoginosins, responsible for the symptoms of the disease.

Toxic diatoms

After consumption of cultivated mussels an outbreak of food poisoning occurred due to a glutamate antagonist in central nervous system, called domoic acid, produced by Nitzschia pungens (20) (21), a chain-forming diatom of the phytoplankton.

CONCLUSIONS

The clinical symptoms of many gastrointestinal disorders caused by parasites remain usually similar regardless of the etiological agent of the disease. The symptoms of an intestinal infection range from the very mild (most common) to an acute or life-threatening disease. Other non bacterial agents of foodborne disease are toxigenic fungi or algae, which can produce very toxic compounds, causing not only gastrointestinal symptoms but often very serious life-threatening disease states.

ACKNOWLEDGEMENTS

This review has been carried out with financial support from the Commission of the European Communities, Agriculture and Fisheries (FAIR) specific RTD programme PL98 4230 'Intestinal Flora: Colonisation Resistance and Other Effects'. It does not necessarily reflect its views and in no way anticipates the commission's future policy in this area.

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