REVIEW

Extra-intestinal and long term consequences of *Giardia duodenalis* infections

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Abstract

Giardiasis is the most common waterborne parasitic infection of the human intestine worldwide. The etiological agent, *Giardia duodenalis* (syn. *G. intestinalis*, *G. lamblia*), is a flagellated, binucleated protozoan parasite which infects a wide array of mammalian hosts. Human giardiasis is a true cosmopolitan pathogen, with highest prevalence in developing countries. Giardiasis can present with a broad range of clinical manifestations from asymptomatic, to acute or chronic diarrheal disease associated with abdominal pain and nausea. Most infections are self-limiting, although re-infection and chronic infection can occur. Recent evidence indicating that *Giardia* may cause chronic post-infectious gastrointestinal complications have made it a topic of intense research. The causes of the post-infectious clinical manifestations due to *Giardia*, even after complete elimination of the parasite, remain obscure. This review offers a state-of-the-art discussion on the long-term consequences of *Giardia* infections, from extra-intestinal manifestations, growth and cognitive deficiencies, to post-infectious irritable bowel syndrome. The discussion also sheds light on some of the novel mechanisms recently implicated in the production of these post-infectious manifestations.

Core tip: This review offers a state-of-the-art discussion on the long-term consequences of *Giardia* infections, the most common waterborne parasitic infection of the human intestine worldwide, from extra-intestinal manifestations, growth and cognitive deficiencies, to post-infectious irritable bowel syndrome. The discussion also sheds light on some of the novel mechanisms recently implicated in the production of these post-infectious manifestations.

INTRODUCTION

Gardia duodenalis (*G. duodenalis*) (syn. *Giardia lamblia*, *Giardia intestinalis*) is an intestinal flagellated protozoan parasite of the upper small intestine. Very common worldwide, *Giardia* was recently included in the World Health Organisation's Neglected Disease Initiative[1,2]. *Giardia* is

Table 1 Main pathophysiological effects of *Giardia duodenalis* and their mechanisms

Giardia-induced pathophysiological responses	Mechanisms involved or hypothesized to be involved	Selected references
Intestinal epithelial cell apoptosis	Induction of pro-apoptotic factors: Caspase-3, 8 and 9, Inhibition of anti-apoptotic factors: poly (ADP-ribose) polymerase (PARP) cleavage	[10,18,19,21,23]
Halt of enterocyte cell-cycle progression	Nutrient competition (arginine), up-regulation of cell-cycle genes	[25]
Intestinal barrier dysfunction	Disruption of claudin-1 and alpha-actinin by unknown mechanisms, caspase-3 mediated disruption of zonula-occludens (ZO)-1, myosin light chain kinase-mediated disruption of F-actin, and ZO-1	[10,17,19,21,26,27,29,30]
Small intestinal hypermotility	Adaptive immunity, neuronal nitric oxide, mast cell degranulation	[118-120]
Diffuse shortening of brush border microvilli	CD8+ lymphocytes - mediated *via* parasite secretory/excretory products	[13,16,21,31]
Crypt hyperplasia	Alteration villus/crypt ratio	[21,62,99]
Microbiota composition	Microbiota from infected host may become pathogenic	[14,33]
Increased mucus production	Increased mucus secretion in response to the parasite	[66]
Brush border enzyme activity deficiencies	Loss of surface area (microvilli and villi)	[16,21,98,121,122]
Disaccharidases deficiencies	Loss of surface area (microvilli and villi)	[10,16,99,122]
Electrolyte/nutrient/water malabsorption	Loss of surface area (microvilli and villi)	[10,19,21,62,99,123]
Anion hypersecretion	Unknown mechanisms	[10,19,99]

PARP : Poly adenosine diphosphate ribose polymerase.

transmitted through the ingestion of cysts in contaminated food or water, or directly *via* the fecal/oral route. Ingestion of cysts results in giardiasis, a disease causing intestinal malabsorption and diarrhea in a wide variety of species including humans. In developing countries, the prevalence of human giardiasis commonly ranges from 20% to 30% of the population, with reports of 100% prevalence in some populations; in developed countries, prevalence ranges from 3% to 7%[3,4]. The classification of *G. duodenalis* is a topic of debate and at present, the species is divided into eight distinct genetic assemblages, *i.e.*, assemblages A to H. Only the assemblages A and B are considered to be pathogenic in humans. Although parasites with assemblage A or B can infect non-human mammalian species, other genotypes appear to have a more restricted host range; for example assemblages C and D are commonly found in dogs[5], while assemblage E is common in cattle[6]. Ongoing research suggests that giardiasis is often due to anthroponotic spread, but zoonotic transmission can occur[7-9]. A striking feature of giardiasis is the spectrum of clinical symptoms that occur in infected individuals. The clinical manifestations can range from asymptomatic, to acute or chronic diarrheal disease. When present, the clinical signs of infection may include diarrhea, nausea, weight loss, bloating and abdominal pain[3,10]. In giardiasis, the acute pathophysiology occurs without invasion of the small intestinal tissues by the trophozoites, and in the absence of overt inflammatory cell infiltration, with the exception of a modest increase in intraepithelial lymphocytes[11-13]. Multiple factors have been proposed to account for the disease variability, including the state of the host immune system, host age and nutritional status, strain genotype, infectious dose and, possibly co-infections[3,8,10]. The pathophysiological consequences of *Giardia* infection are clearly multifactorial, and involve both host and parasite factors, as well as immunological and non-immunological mucosal processes. Recent observations suggest a role for disruptions of the host intestinal microbiota during the acute infection stage in the production of chronic symptoms, and further research is warranted to corroborate these findings[14]. The pathophysiology of giardiasis, and key aspects of the host response to *Giardia* remains incompletely understood.

PATHOPHYSIOLOGY OF GIARDIASIS

Central features of the pathophysiology of giardiasis are briefly outlined below, as these mechanisms may be key to our understanding of the complications discussed further (Table 1). While the *Giardia* genotype has been proposed to play a role in the induction of symptoms, there is currently no consensus concerning the connection between genotype and virulence[15].

After cyst ingestion in contaminated water or food, excystation occurs liberating two or four trophozoites, which adhere to the epithelial surface of the intestine *via* a ventral adhesive disk. This tight attachment between *Giardia* trophozoites and intestinal epithelial cells, as well as the production of yet incompletely characterized parasitic products, culminate in the production of diarrhea. Pathophysiology is believed to involve heightened rated of enterocytes apoptosis, intestinal barrier dysfunction, activation of host lymphocytes, shortening of brush border microvilli with or without coinciding villous atrophy, disaccharidase deficiencies, small intestinal malabsorption, anion hypersecretion and increased intestinal transit rates[10,13,16-22].

As it is the case with other enteropathogens, induction of apoptosis in enterocytes by *Giardia* represents a key component in the pathogenesis of the infection[3,18,19,22-24]. Enterocytes apoptosis during giardiasis is caspase-3 and -9 dependent[18,23]. While both host and parasite factors may modulate intestinal epithelial cell apoptosis, the products responsible for its activation during giardiasis have yet to be identified. In addition to promoting increased rates of enterocyte apoptosis, *Giardia* trophozoites may also halt enterocyte cell-cycle progression *via*

consumption of arginine, and up-regulation of cell-cycle inhibitory genes[25].

Findings from studies on giardiasis *in vivo* demonstrate that the most severe intestinal permeability and macromolecular uptake coincides with the peak of trophozoites colonization[17,26-28]. The effects of the infection on gut barrier function following host parasite clearance require further investigation. *Giardia*-mediated increases in intestinal permeability result from alterations to the apical tight junctional complexes, including disruption of F-actin, zonula-occludens-1, claudin-1 and alpha-actinin, a component of the actomyosin ring that regulates paracellular flow[19,26,28-30]. The role of *Giardia* proteinases in these effects is a topic of ongoing research.

Giardia-induced diffuse shortening of epithelial brush border microvilli represents a key factor in the production of diarrhoeal disease *via* malabsorption and maldigestion[13,16,31]. Whether or not the diffuse loss of microvillous border surface area associated with giardiasis is related to the release of a "toxin" by the parasite, a phenomenon similar to the release of proteases in the bacterial overgrowth syndrome[32], remains poorly understood. Regardless, *G. duodenalis* infection causes microvillous shortening in a lymphocyte-mediated manner which in turns impairs activities of disaccharidases[13].

Bacterial components of the intestinal microbiota from *Giardia*-infected hosts may act as stimulatory factors for protozoan pathogenicity[33]. Indeed, micro-organisms isolated from the duodenal microbiota of patients with symptomatic giardiasis can stimulate the pathogenicity of *G. duodenalis* in a gnotobiotic animal model[33]. The biological basis of this phenomenon remains unclear.

Giardia infections tend to be self-limiting in individuals with competent immune systems. A recent study in Brazilian children suggests that symptoms are less severe during re-infection, consistent with the hypothesis that if previous exposure does not always protect against future infections, it does at least reduce the severity of pathology[34]. Patients with common variable immunodeficiency and Bruton's X-linked agammaglobulinemia are prone to chronic giardiasis[35,36], which underscores the necessity of antibodies to fully control giardiasis.

In addition to its acute symptoms, giardiasis may also cause anorexia and failure to thrive. Indeed, *Giardia* infections may have detrimental effects on nutritional status, growth status and cognitive function in humans[37-41]. *Giardia* infections may also have detrimental effects on body weight in food-producing animals making this a serious concern for the agricultural industry[42-45].

LONG-TERM CONSEQUENCES OF GIARDIASIS

Extra-intestinal consequences of Giardia infections

Until recently, the scientific literature rarely reported extra-intestinal manifestations in giardiasis. However, a recent study estimated that 1/3 of the patients infected with this parasite will express long-term extra-intestinal symptoms, suggesting that this phenomenon is not as uncommon as previously thought[46].

Ocular pathologies: The first description of ocular complications in patients with giardiasis was made by Barraquer[47], who reported cases of iridocyclitis, choroiditis, and retinal hemorrhages in patients that presented diarrhea linked to the presence of *Giardia*. More recent observations described a "salt and pepper" degeneration (punctuate areas of normal hyperpigmentation on a light yellow pink-retina) involving the retinal pigmented epithelium in children suffering from giardiasis[48]. The same complication was described in children with past giardiasis, indicating that the ocular changes observed did not require the concurrent presence of the parasite in the gut[49]. Small children appear to be more susceptible to ocular lesions during giardiasis, and the lesions are thought to be caused by damage to the cells of the retina, accompanied by the release of pigment granules in retinal layers, where they can be seen as blackish dots[49]. The mechanisms linking ocular lesions with giardiasis remain obscure, but they exclude the possibility of direct invasion by the parasite. It has been speculated that the pigmented degeneration may result from toxic metabolites produced by the parasites, which has yet to be proven[48]. The role of increased intestinal permeability in the ocular complications seen in giardiasis needs to be elucidated.

Arthritis: Reactive arthritis is classically seen following infection with enteric pathogens such as *Yersinia* sp*., Salmonella* sp*., Campylobacter jejuni* and *Shigella* sp., but inflammatory arthritis has also been described following enteric infections with other organisms such as *Clostridium difficile, Brucella* sp. and *Giardia* sp.[50]. The interval between the preceding infection and the manifestation of arthritis is 2 to 4 wk[50]. Post-infectious arthritis has a predilection for joints of the lower limbs particularly the knee and ankle[51]. Post-infectious reactive arthritis has been classified as a classical spondyloarthropathy associated with human leukocyte antigen (HLA)-B27, an allele of the major histocompatibility complex class I present in 50% of the cases of patients with enteric-infection-related arthritis[51,52]. However, inflammatory arthritis following infection with *Clostridium* sp. or *G. duodenalis* does not fit classical spondyloarthropathy, as it fails to show association with HLA-B27[50]. Therefore, these are referred to enteric-infection-related-*arthritides*. Although *G. duodenalis* infections account for a significant proportion of enteric infections worldwide, reports of an association with post-infectious arthritis are relatively few. Little is known of the pathogenesis of arthritis in these conditions. Unlike post-enteric reactive arthritis, these arthritides are characteristically responsive to antibiotic therapies[52]. The variable degrees of host immune responses, and the lack of a robust systemic inflammatory response, may account for the infrequency of post-giardiasis arthritis despite the high prevalence rate of the infection[50]. Antigens from enteric bacteria have been isolated from the synovial fluid

Table 2 International reports of post-giardiasis metabolic consequences		
Post-giardiasis effects	**Country**	**Selected references**
Lower cognitive function	India, Peru, Turkey	[40,67,68,76,77]
Lower intellectual quotient		
Lower social quotient		
Lower weight	Brazil, Columbia, Ecuadora,	[37-39,63,64,66-68,72,77,78,124-129]
Lower height	Guatemala, Iran, Israel,	
Stunting	Mexico, Rwanda, Turkey, United States	
Failure to Thrive	Columbia, Ecuadora, United States	[64,66,127]
Nutrient deficiencies	Iran, Mexico, Tanzania	[38,69,78,81]

of affected joints[52]. In a case of *Yersinia pseudotuberculosis* reactive arthritis, evidence of viable bacteria within the joint was demonstrated over a year later[53]. Here again, a possible role for increased intestinal permeability in enteric-infection-related-arthritis warrants further investigation.

Allergies: Concomitant presence of *G. duodenalis*, cutaneous allergic manifestations, and gastrointestinal symptoms have been described, which may explain why complete symptom resolution can be achieved with metronidazole and corticosteroids[54]. Significant anecdotal evidence suggests a causative link between giardiasis and the development of urticaria. In a recent study in children, giardiasis was associated with an increase in total serum immunoglobulin E (IgE) levels, and an enhanced IgE antibody response to common allergens[55]. These patients also demonstrated IgE reactivity to cow's milk and *Giardia* antigens. These observations suggests that alteration in antigen uptake from the small intestine during giardiasis, perhaps in association with connective tissue mast cell proliferation, may contribute to the development of allergic disease[56-58]. Dysfunction of the intestinal barrier during giardiasis may facilitate the translocation of food macromolecules and in turn prime the host for sensitization[55].

Muscular complications: Hypokalemic myopathy has been associated with celiac disease, radiation enteropathy, immunosuppressive drugs, and various infectious diseases. In the patient, this presents as marked proximal muscular weakness in all four limbs and the neck[59]. Analyses of muscular biopsies reveal an abnormal size of the muscular fiber due to the presence of numerous rounded atrophic and hypertrophic fibers, proliferation of myonuclei, and necrotic fibers[60]. The findings are consistent with impairment of muscle excitability and denervation due to muscle necrosis. Analysis of these fiber components showed that glycogen and lipid levels, as well as the inter-myofibrillar network pattern, are normal[60]. Several cases of myopathy following hypokalemia induced by giardiasis have been reported in both immunocompetent and immunocompromised patients[59,60]. This suggests

that *G. duodenalis* infections can trigger muscular manifestations independently of the immune status of the host. During giardiasis, potassium loss is closely related to the number of bouts of diarrhea per day[60]. Loss of potassium result in hypokalemia which can trigger a severe and transient myopathy[60]. In fact, muscular symptoms can improve with increased levels of potassium and recovery from diarrhea[59]. However, *G. duodenalis* diarrhea as a cause of myopathy due to hypokalemia is rare. It seems that the duration of symptoms is crucial for development of hypokalemic myopathy[60]. Giardiasis-associated hypokalemia occurs more often in elderly people, particularly women, who are hospitalized for giardiasis[61]. The causes, and the clinical consequences, of *Giardia*-associated hypokalemia remain unclear. It has been suggested that giardiasis-induced impairment of nutrient and electrolyte absorption may contribute at least in part to hypokalemia and hyponatremia[62].

"Metabolic" consequences of Giardia infection

Nutritional consequences: In developing countries of the world, because of infectious diseases and lack of food, 206 million children under 5 years of age suffer from stunting, 50 million from chronic wasting disease, and 167 million are grossly underweight[63]. Growth failure, reflected in stunting, wasting and underweight conditions, is assessed by anthropometric indices of height-for-age, weight-for-age, and weight-for-height[64]. Optimum health for children has long been linked to physical, socio-cultural, economic and environmental factors. In developing countries, the incidence of giardiasis is often over four times higher than the incidence reported in industrialized countries[65]. Children between 6 mo and 5 years of age are the most susceptible[66]. In combination with diarrhea, *G. duodenalis* infection can cause iron deficiency anemia, micronutrient deficiencies, protein-energy malnutrition, growth and cognitive retardation, and malabsorption[63,67]. Studies conducted on children from Brazil and Peru found that diarrheal disease occurring in the first 2 years of life negatively correlates with cognitive function, verbal fluency, and physical fitness, and may lead to long-term growth faltering[40,68]. These studies demonstrate that the effects of early childhood diarrhea are more far-reaching than merely causing dehydration. Diarrhea caused by *Giardia* sp. or *Cryptosporidium* sp. has frequently been associated with stunting and lower cognitive function[40,68] (Table 2). Intriguingly, a recent study observed that in a cohort of Tanzanian children infected with *Giardia*, infection had a protective role against diarrhea, and that this protection was lost with multi-nutrient supplementation[69]. Research needs to determine whether these interesting findings reflect a negative regulation by *Giardia* sp. of other enteric pathogenic processes that may occur in these children.

Failure to thrive: Childhood and adolescence are the period of most rapid skeletal growth. Failure to thrive (FTT) is a term generally used when a child presents

with a rate of weight gain that is significantly below that expected of similar children of the same sex, age and ethnicity. Failure to thrive is a common problem, that may be present at any time during the childhood, but is usually prevalent within the first 1-2 years of life. Long-term sequelae involving all areas of growth, behaviour and development may be seen in children suffering from FTT[70]. Causes for FTT usually include: (1) inadequate food intake; (2) reduced absorption or digestion of nutrients or excessive loss of nutrients; and (3) excessive utilisation of energy. There is a strong association between *Giardia* infection and malnutrition, wasting and stunting[38,63-65,69,71]. Malabsorption, maldigestion and malnutrition due to giardiasis have been shown to affect anthropomorphic factors as well as the calorie intake during childhood, most commonly in the second year of life in infected children[37-39,63,72]. Duration of the infection episodes, and their association with diarrhea, appeared to be the key factors associated with growth disturbance and failure to thrive[37]. While several studies have established a strong link between *Giardia* infection during the first two years of life, FTT and development impairment, more research is needed to unravel the mechanisms and the potential implications of polyparasitism in these phenomena.

Malnutrition, a common feature of numerous intestinal diseases, has been associated with an increase in macromolecular uptake due to heightened intestinal permeability[73], two phenomena known to occur during giardiasis[19,56]. *Giardia* infection can reduce food intake, and produce steatorrhea, maldigestion and malabsorption of carbohydrates and vitamins (including vitamin A, B3, B5, B6, B12, E, and folacin)[3,21,64,74]. Together, these effects may contribute at least in part to failure to thrive in giardiasis (Table 2).

Stunting: Growth failure due to malnutrition and chronic infections like giardiasis is associated with increased morbidity and mortality in children from developing countries[37,63,64]. More specifically, significant impairments in weight-for-age and weight-for-height scores have been associated with *G. duodenalis* infection during the first two years of life[72]. Indeed, the relative odds of low height-for-age may be 7.7 times higher among children with giardiasis[63]. In a number of developing countries, diarrhea caused by enteric parasitic Protozoa in early childhood represents predictors of stunting[64]. Given the high prevalence of asymptomatic infection in this study population (78.8%), children may appear to have normal weight-for-age and weight-for-height early on, but, present with growth retardation at a later age. This phenomenon is known as "homeorhesis", and it is probable that the high prevalence of asymptomatic *Giardia* infection among children may play a key role in it. Similarly, *Giardia* infection has been associated with decreased weight gain and impaired feed conversion efficiency in lambs and cattle, illustrating that growth retardation associated with *Giardia* infections also poses an important problem to the agriculture industry[42-45]. Overall, human giardiasis combines with other factors, including low nutritional status, as well as sanitary and socioeconomic conditions, to lead to stunting[64]. However, findings from numerous studies, to date, indicate that the well established loss of intestinal surface area, maldigestion, and malabsorption caused by giardiasis may contribute to growth retardation following *Giardia* infection (Table 2).

Impaired cognitive function: Cognitive function in children can be affected by environmental and health related factors[75]. Risk factors that interfere with cognitive function are especially important during infancy because the first two years of life are an essential period of rapid growth and development, that is marked by rapid brain growth and maturation, by neuronal arborisation, myelinisation and emergence of brain networks. Thus the development of cognitive function in early life depends on the hierarchical maturation of neocortical association areas, as well as interactions with the environment. Nutrition, infection, and environment, have been found to affect neuroplasticity and to have long lasting effects in developing children[76]. Many of the hazards to early brain development are well known, and include head injury, newborn asphyxia, infections of the brain *in utero* and in the first year of life, genetic defects, lead poisoning and malnutrition. Micronutrient deficiencies (*e.g.,* Iodine) and iron deficiencies have also been found to impair cognitive development[76]. Studies have attempted to link possible long-term cognitive deficits with severe diarrhea in early childhood[40,41,68]. The complex interrelation among malnutrition, diarrheal disease and environmental factors such as socioeconomic status and education make it difficult to determine the unique contribution of either malnutrition or diarrheal disease to cognitive development. However, chronic malnutrition and stunting during infancy secondary to *G. duodenalis* infections, has been associated with poor cognitive function[40,41,77]. Moreover, diarrhea during early childhood was also found to impair visual-motor coordination, auditory short-term memory, information processing, and cortical cognitive function[68,76].

Interestingly, poor language cognition and impaired psycho-motor development appear to be associated with *Giardia* sp. more so than with other enteropathogenic parasites such as *Entamoeba histolytica, Ascaris lumbricoides, Enterobius vermicularis,* or *Trichuris trichiura*[63]. These studies have suggested a role for nutrient malabsorption and micronutrient deficiencies, such as zinc, iron or vitamins (A and B-12) in humans as well as in animals[63,74,78,79]. Indeed, significantly lower levels of iron and ferritin, known to affect psychomotor development, have been detected in patients with giardiasis[64]. Similarly, diarrhea due to giardiasis was linked to poor cognitive function by causing zinc and iron micronutrient deficiencies, as well as defects in the anti-oxidant system, which may all affect neuroplasticity[76]. Indeed, perinatal iron deficiency in rats reduces neuronal metabolic activity, specifically targeting

areas of the brain involved in memory processing[80]. Zinc supplementation was recently found to reduce the rate of diarrhea caused by giardiasis[81]. The complexity of these profound effects on functional impairments requires further investigation. More research is also needed to determine whether and how these effects can be reversed with targeted antimicrobials, with micronutrient and/or oral rehydration, or nutrition therapy[68] (Table 2).

Chronic fatigue syndrome: Viral, bacterial, as well as parasitic pathogens can trigger chronic fatigue syndrome (CFS), and are responsible for work-related disability reflected in long-term sickness, absence from studies and employment[82]. Although the biological basis of CFS is unknown, it is generally thought that post-infectious fatigue develops shortly after acute infection. CFS has been described following Q-fever, Epstein-Barr virus infection, Ross river virus infection, brucellosis, Lyme disease, viral meningitis and Dengue fever[83]. Recent studies have reported a high prevalence of post-infectious fatigue following a giardiasis outbreak in Bergen, Norway, in 2004[15,82-86]. Fatigue was reported in 41% of the people in Bergen 2 years after the *Giardia* outbreak, compared to 22% in the general population[82]. In this population, old age and female gender were a significantly higher risk factor for post-infectious fatigue[84,87].

Although *Giardia* is a non-invasive parasite, post-giardiasis CFS is likely to include immunologic components[82]. Studies have implicated differences in activation and function of peripheral blood lymphocytes subsets in post-giardiasis CFS, including altered natural killer-cell levels and lowered CD4:CD8 ratios[87,88]. The exact roles of immune factors in co-morbidities associated with gastrointestinal disorders and CFS need to be further explored. Fatigue is a frequent symptom in patients with functional gastrointestinal disorders (FGID), especially irritable bowel syndrome (IBS)[89].

Chronic gastrointestinal consequences of Giardia infections

FGID: FGID represent a group of disorders characterized by recurring or chronic gastrointestinal symptoms without an identifiable disease process. IBS and functional dyspepsia (FD) are the best described FGID. Post-infectious-IBS (PI-IBS) has been reported following acute gastroenteritis due to bacteria such as *Salmonella* sp., *Shigella* sp. and *Campylobacter jejuni*[90,91]. Recent evidence now indicates that a proportion of patients diagnosed with *Giardia duodenalis* will also develop PI-IBS symptoms in the absence of detectable parasitic loads[92,93].

Post-infectious irritable bowel syndrome: IBS is the most common functional gastrointestinal disorder diagnosed by gastroenterologists today. It is characterized by abdominal discomfort and altered bowel habit, with no abnormality on routine diagnostic tests. Several mechanisms have been considered in the pathogenesis of IBS including genetic, psychological and environmental factors as well as intestinal motor and sensory func-

tions associated with brain-gut interactions[94]. In some patients, IBS symptoms seem to arise *de novo* following acute gastroenteritis (GE). This PI-IBS denotes the persistence of abdominal discomfort, bloating and diarrhea, despite clearance of the inciting pathogen. Recent meta-analyses demonstrated that the risk of developing IBS increases six-fold after gastrointestinal infection and remains elevated for at least 2-3 years post-infection, and it is estimated that 7%-31% of patients with infectious GE go on to develop PI-IBS[90,91]. Higher risk factors include longer duration of symptoms, young age and female gender. The current conceptual framework regarding the pathophysiological mechanisms for PI-IBS suggests that it is associated with increased intestinal permeability and motility, increased numbers of enterochromaffin cells and persistent intestinal inflammation, characterized by increased numbers of T-lymphocytes and mast cells, and increased expression of pro-inflammatory cytokines[95-97]. Possible mechanisms for PI-IBS include genetic predisposition, motility dysfunction, such as accelerated colonic transit and smooth muscle hyper-reactivity to acetylcholine, continuous antigenic exposure (bacterial, parasitic or dietary), or molecular mimicry of foreign antigens[98].

Early reports indicated that *Giardia* may cause prolonged symptoms, including secondary lactose intolerance, for several weeks after successful treatment[99]. Chronic giardiasis resembles IBS, and symptomatic infection may exacerbate existing IBS[100]. *Giardia* infection has been diagnosed in 5%-10% of patients with IBS[101,102], and it was recently demonstrated that *G. duodenalis* may indeed cause IBS and functional dyspepsia[93]. High frequency of microscopic duodenal inflammation was found in patients post-giardiasis when the infection lasted 2-4 mo, further supporting the hypothesis that longer duration of infection is a risk factor for PI-IBS[103]. Early diagnosis of *Giardia* infection and treatment may shorten the duration of the infection and hence may help reduce the risk for such complications[83].

Interactions between the host and gastrointestinal microbiota may play a key role in the pathogenesis of IBS. Fecal microbiota are altered in patients with IBS, and patients with diarrhea-predominant IBS appear to host more *Proteobacteria*, and fewer *Bacteroidetes* compared to asymptomatic patients[104,105]. The mechanisms by which altered fecal flora may induce disease are poorly understood. Abnormalities in short chain fatty acids have been reported in patients with diarrhea-predominant IBS[105]. Whether these alterations may result from abnormalities in the host microbiota requires further investigation[14].

Historically, IBS was considered as a psychosomatic disorder, with an emphasis on psychiatric comorbidity[106,107]. During the past decades, GE and low grade inflammation as mechanisms underlying gastrointestinal (GI) dysfunction have been involved in IBS symptoms[106,108]. It is now well established that there is a relationship between the neural and immunological networks within the gut, and that the central nervous system and the gut are engaged in constant bi-directional communication, often related to as the brain-gut axis (BGA).

Table 3 Extra-intestinal and long-term consequences of giardiasis

Post-infectious consequences	Speculated mechanisms involved	References
Ocular pathologies	Speculated involvement of toxic metabolites produced by the parasite	[47-49]
Arthritis	Bacterial antigens in synovial fluids possibly due to increased intestinal permeability	[50-52]
Allergies	Alteration in antigen uptake	[54-57]
	Dysfunction of the intestinal barrier	
Hypokalemic myopathy	Loss of potassium related to diarrhea, impaired nutrient and electrolyte absorption	[59-62]
Failure to thrive	Inadequate food intake, Reduced nutrients absorption, excessive utilisation of energy, Steatorrhea, maldigestion, malabsorption	[38,39,63-65,69,71,118]
Stunting	Nutritional status, sanitary, socio-economic conditions, loss of intestinal surface area, maldigestion, malabsorption	[37,63,64,67,77,121]
Impaired cognitive functions	Chronic malnutrition and stunting following *G. duodenalis* infection	[40,41,63,64,68,76-78]
	Nutrient malabsorption and micronutrient deficiencies	
Chronic fatigue syndrome	Altered natural killer-cell levels	[15,82-87,89]
	Lower ratio CD4:CD8	
Post-infectious irritable bowel syndrome	Microscopic duodenal inflammation	[14,84,93,100-105]
	Interaction host - gastrointestinal microbiota	
	Increased T-cells and Mast-cells	
Cancer	No cause-to-effect established	[113-116]

Among the pathophysiological mechanisms of IBS, disorder of the BGA has been associated[106,108]. Recently, more evidence of emerging dysbiosis in IBS patients have been made[104], suggesting an important role of the microbiota-gut-brain axis[106-111]. Nevertheless, our understanding of the mechanisms of the bi-directional interactions between microbiota and GI physiology and its association with behavior needs to be explored with focus on the contributions of immunological and neural components to the microbiota-BGA relationship. Insights into the interactions between enteric pathogens, the host epithelia, and the intestinal microflora are needed to improve our understanding of disease processes that may initiate IBS or even exacerbate intestinal inflammation in patients with IBD[112]. Studies on giardiasis offer a powerful model to investigate these mechanisms.

Cancer

A few reports have described *Giardia* trophozoites in the tumoral mass of pancreatic tissue and gallbladder. While *G. duodenalis* trophozoites are generally localized to the proximal small bowel, they may also be identified in the stomach, distal small bowel, or caecum, and studies have reported pancreatic infection with *Giardia*[113-115]. Although the relationship between pancreatic giardiasis and pancreatic cancer is presently unknown, the coexistence of these 2 diseases may prompt exploration into mechanisms of carcinogenesis in giardiasis. In another study, following cholecystectomy with liver bed resection and lymph node dissection, intra-operative cytological examination of the patient's bile juice revealed the presence *G. duodenalis* trophozoites, and pathological examination revealed gallbladder cancer[116]. However, no cause-to-effect has yet been established between the presence of *Giardia* and the development of cancer.

CONCLUSION

Infections with *Giardia duodenalis* may remain asymp-tomatic, or cause acute or chronic diarrheal disease. The observations discussed herein also demonstrate that, in addition to its classical intestinal presentation, giardiasis may cause ocular complications, arthritis, skin allergies or myopathy. Moreover, giardiasis is now a well established cause of failure to thrive, stunting and growth retardation in human and animals, diminished cognitive functions, and chronic fatigue. Finally, *Giardia* may lead to post-infectious functional gastrointestinal disorders such as irritable bowel syndrome and functional dyspepsia. A few cases of *Giardia* trophozoites associated with tumoral masses have also been reported, but cause-to-effect relationships between giardiasis and cancer have yet to be established (Table 3).

Long-term complications of giardiasis may present 2 to 3 years following the infection. In some cases, they may last for a few weeks, and may be eliminated with anti-parasitic treatment, observations that have been reported for example in cases of myopathy and skin allergies. In other cases, long-term consequences may be present for several years, in the form of failure to thrive, stunting, IBS, and chronic fatigue syndrome, in the absence of any parasite.

The mechanisms responsible for post-infectious and extra-intestinal manifestations in giardiasis remain obscure. Both parasitic and host factors have been implicated, indicative of a multifactorial pathogenic process. However, as anti-microbial treatment often leads to recovery, the infection itself represents a key element in these complications. As giardiasis can be asymptomatic, the complex processes leading to extra-intestinal and post-infectious manifestations represent a challenging topic for further research (Table 3).

Given the high prevalence of giardiasis in young children in developing countries, its significant effects on stunting and wasting, and the lasting effects of early childhood diarrhea and malnutrition, giardiasis is of considerable public-health importance. Even though the consequences of giardiasis are variable, school health

programs and health education for children and parents aimed at reducing the prevalence of intestinal parasitic infection in children may have beneficial effects on child growth and development. Improved diagnostic methods, particularly in asymptomatic patients, as well as more-effective treatment and control strategies, are sorely needed to help reduce the detrimental impact of the infection on human societies as well as in the agriculture industry.

In the recent few years, and particularly since the 2004 giardiasis outbreak in Bergen, Norway, *G. duodenalis* infections have been linked to post-infectious IBS and functional dyspepsia *via* mechanisms that are unclear. Findings from several studies indicate roles of specific pro-inflammatory cytokines, and hyperlplasia of enterochromaffin cells, mast cells, and lymphocytes, perhaps causing motility dysfunction and visceral hypersensitivity; but much controversy remains on the topic[117]. Together, the data strongly suggest that the appearance of post-infectious complications in giardiasis are multifactorial. A number of the post-infectious complications seen after giardiasis are shared with those caused by common bacterial enteropathogens like *C. jejuni*, *E. coli*, or *Salmonella* sp. One area of growing interest in this field is the role played by disruptions of the host microbiota during the acute stage of the infection in the initiation of delayed immune-mediated pathophysiology. Therefore, a better understanding of the mechanisms responsible for the extra-intestinal and post-infectious manifestations of giardiasis will help unravel common pathways leading to these phenomena.

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