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Giardiasis – why do the symptoms sometimes never stop?

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Although giardiasis is considered by most medical practitioners to be an easily treated infection, prolonged symptoms due to, or following, *Giardia duodenalis* infection can have a significant impact on quality of life. Symptom recurrence, including abdominal symptoms and fatigue, can result from re-infection, treatment failure, disturbances in the gut mucosa or post-infection syndromes. In developed countries, these sequelae can have an enormous impact on quality of life; in developing countries, particularly in children, they add yet another burden to populations that are already disadvantaged. Here, we outline current knowledge, based on individual case sequelae from sporadic infections, observations of population effects following outbreaks and studies of phenotypic and genotypic diversity between morphologically identical isolates of parasites. We also raise further questions, looking for clues as to why giardiasis sometimes becomes an intrusive, long-term problem.

Giardiasis: a re-emerging infectious disease

Over 320 years since the aetiological agent of giardiasis was first observed by van Leeuwenhoek, *Giardia duodenalis* (syn. *G. intestinalis*, *G. lamblia*) continues to be one of the most common intestinal parasitic protozoa reported in humans, worldwide. The parasite also infects a wide range of other mammalian hosts, including livestock, cats, dogs, rodents and artiodactyls. Molecular studies have divided this species into various assemblages or genotypes, which not only demonstrate host specificity patterns (only assemblages A and B are zoonotic, infectious to humans and various other mammals), but also differ in a range of other phenotypic aspects [1]. Whether these variations are sufficient to result in a reorganisation of the current taxonomy of *Giardia* is under debate (https://community.eupathdb.org/).

Transmission of *Giardia* is via the faecal–oral route, either indirectly through contaminated water or food, or directly from person to person. Often predominantly associated with developing countries, where compromised hygiene infrastructure might lend itself to increased transmission and endemic establishment of such diseases, giardiasis has been included in WHO's 'Neglected Disease Initiative' since 2004 [2]. In industrialised countries, its role in outbreaks of diarrhoeal disease in day-care centres and water-associated outbreaks has resulted in giardiasis being sometimes referred to as a re-emerging infectious disease [3].

Signs and symptoms

Giardia infection is usually associated with diarrhoea, but can be either asymptomatic or responsible for a broad clinical spectrum, with symptoms ranging from acute to chronic [4]; diarrhoea can occur with or without malabsorption syndrome; there can be nausea, vomiting, and weight loss [5]. Occasionally, *Giardia* infection can be associated with pruritis and urticaria [6], uveitis [7], sensitisation towards food antigens [8,9] and synovitis [10]. Children might also suffer more serious consequences, including retarded growth and development [11,12], poor cognitive function [13] and detrimental effects on nutritional status [13–15]; however, the latter is considered to be controversial, as another research study has not demonstrated this effect [16]. Giardiasis could be self-limiting in some cases, but because of the potential for chronic or intermittent symptoms, treatment is recommended. At least six different classes of drugs, with different mechanisms, are available for giardiasis treatment (Table 1) [17], but 5-nitroimidazole compounds are usually the agents of choice. The alternatives could be used if other advantages are appropriate or if 5-nitroimidazole therapy fails.

The spectrum of symptom patterns and the occurrence of treatment failures have long been recognised, but in an outbreak situation (such as in the Bergen outbreak [18] see below) can be brought sharply into focus. Some patients have a mild, inconsequential illness that resolves spontaneously or responds immediately to treatment with a 5-nitroimidazole compound. Others suffer a severe, long-lasting illness, for which treatment is ineffectual, and, even after the parasite has finally been eliminated, some sequelae persist, affecting quality of life, and continuing to cause the patient discomfort or pain. Most patients are somewhere between these two extremes. Those who experience prolonged, symptomatic giardiasis can feel driven almost to despair. In this article, we attempt to address their question, why do the symptoms sometimes never stop?

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Table 1. Current antigiardial agents

Antigiardial agents	Efficacy[a] (%)	Adverse events reported	Effective dosage		Other comments
			Adults	Children	
Five-nitroimidazole compounds		Gastrointestinal discomfort, metallic taste, disulphiram-like effects. Headache, vertigo, insomnia, irritability, neuropathy, seizures. Rash. Reddish-brown urine. Transient elevation of transaminases Leukopenia.			First choice of treatment.
Metronidazole	36–100		200 mg tid × 7d 500 mg sd × 10d 500 mg tid × 5d	15–20mkd3 × 7d 22.5mkd3 × 5d	Efficacy low when course shorter than 5 days.
Other five-nitroimidazole compounds		Better tolerated than metronidazole.			Single-dose treatment as effective as longer course due to longer half-life.
Tinidazole	74–100		1.5–2g sd × 1d	50mk sd × 1d	Syrup available for children.
Ornidazole	90–100	Rare: Hepatitis and associated cholangitis.	1–2g sd × 1d	20–40mk sd × 1d	
Secnidazole	79–98		2g sd × 1d	30mk × 1d	
Nitrofuran derivatives					
Furazolidone	20–92	Nausea, vomiting, diarrhoea. Haemolysis in G6PD-deficiency. Disulphiram-like activity. Interaction with MAO inhibitors. Haemolytic anaemia in neonates. Brownish urine.	100 mg qid × 10d	6 mkd4 × 10d	Often less effective than other therapies, but has been used in children because of its availability as liquid formulation. Should not be given to neonates or breastfeeding women. Some availability problems.
Benzimidazoles		Usually well tolerated.			Also effective against helminths.
Albendazole	35–96	Nausea, vomiting, diarrhoea, epigastric pain.	400 sd × 5d	10mk × 5 d	Effective in treatment refractory cases in combination with metronidazole.
Mebendazole	42–86	Transient abdominal pain. Potentially severe side effects.	100–200 bid–tid × 1–5d		Optimal dose and duration unclear.
Acridine derivatives					
Quinacrine	84–100	Vomiting, bitter taste, nausea, headache. Yellow discoloration of skin, urine or sclerae (reversible). Urticaria, exfoliative dermatitis, exacerbation of psoriasis. Haemolysis in G6PD-deficiency. Psychosis.	100 tid × 5d	8 mkd3 × 5d	Effective in treatment refractory cases, alone or in combination with other drugs. No longer available in some parts of the world. Possible teratogenic effects.
Amoniglycosides		Usually well tolerated.			
Paromomycin	40–91	Gastrointestinal discomfort.	500 tid × 10d	25 mkd3 × 10d	Clinical data are limited. Recommended for treatment in pregnancy, mainly during the first trimester.
5-nitrothiazolyl derivatives		Usually well tolerated.			Also effective against helminths and other intestinal infections.
Nitazoxanide	64–94	Abdominal pain, diarrhoea, vomiting, headache, yellowish urine.	500 mg bid × 3d	7.5mk bid × 3d	Liquid formulation available for children. Reported effective in a treatment refractory HIV-infected patient.

Other agents *e.g. DL-propranolol, Bacitracin zinc, Chloroquine*	Limited studies.	More studies with greater patient numbers required, but *in vitro* and *in vivo* studies have shown activity for chloroquine and bacitracin zinc; DL-propranolol has been used effectively in combination with metronizadole for cases that are refractory to treatment (reviewed in Ref. [17]).

Abbreviations: sd, single dose; bid, twice a day; tid, three times a day; qid, four times a day; d, days; g, gram; mk, mg per kg; mkd3, mg per kg per day divided into three doses; mkd4, mg per kg per day divided into four doses; G6PD, glucose-6-phosphate dehydrogenase, MAO, monoamine oxidase.

[a]Based on results from clinical studies and reviewed by Escobedo and Cimerman 2007 [17]. Studies vary in design and are not directly comparable. Efficacy based on measuring clearance of the parasite after treatment, but duration of follow up varies between studies

Chronic giardiasis

Chronic giardiasis is not a new concept, and can develop if the infection goes untreated. In Rendtorff's classic infection study in the 1950s, of 14 prison volunteers experimentally infected with *Giardia* cysts, 12 (85%) cleared the parasite spontaneously within 41 days, whereas 2 (15%) were still excreting cysts 146 and 163 days after exposure [19]. In a controlled clinical study of aetiology in malabsorption syndrome in India, a significantly higher number of adult cases (12/50, 24%) had giardiasis, with mean symptom duration of 6.6 months, compared with controls (4/50, 8%) [20]. Lack of detection of cysts from three successive faecal samples is usually used to ascertain absence of infection, but examination of duodenal aspirates or biopsy for trophozoites is also sometimes performed. Limitations in traditional diagnostics in detecting low-level chronic infection should not be overlooked, particularly as such low-level infections, which might be asymptomatic, can contribute synergistically to enhanced pathology in instances of further infection, either with a further strain of *Giardia* or another intestinal pathogen.

Chronic infection is usually associated with diarrhoea and intestinal malabsorption, resulting in steatorrhea, lactase deficiency, and deficiency of vitamin A, vitamin B12 and folate [5]. Epithelial transport and barrier dysfunction are possible mechanisms; in duodenal biopsies from 13 cases with chronic giardiasis, reduced epithelial resistance owing to decreased expression of a tight junction protein (claudin 1) and increased epithelial apoptosis was demonstrated, as well as increased activation of anion secretion and impaired Na^+-dependent D-glucose absorption [21].

Following an outbreak of water-borne giardiasis in Bergen in 2004 [22], with 1262 laboratory-confirmed cases, a prospective cohort study demonstrated that 32% (40/124) of patients with persistent symptoms had chronic *Giardia* infection, with mean disease duration of 7 months [18,23]. Of these, inflammation in duodenal biopsies was found in 87% (34/39); in addition, 54% (21/39) had shortening and blunting of intestinal villi. These cases reported more abdominal pain and diarrhoea than did cases with normal histology [23].

It is probable that there is no single, simple explanation and that the reasons for some cases of giardiasis continuing into chronic infections vary between patients. The two components of the host–parasite relationship might both play a part: 1) host factors including variables such as age, immune status, previous history of exposure, diet and concomitant intestinal microbiota [24]; and 2) parasite factors, probably associated with genotype, including rate of multiplication, variable surface proteins (VSP), resistance to pharmaceuticals and ability to evade immune response. The importance of this interaction between host and parasite characteristics was emphasised among patients with treatment-refractory giardiasis from the Bergen outbreak; although, at the outbreak peak, the *Giardia* isolated from different patients were genetically heterogeneous, they were identical in patients with refractory giardiasis [18]. However, other patients with parasites of the same genetic

make-up (at the genes investigated) responded well to treatment [18]. Thus, both parasite and host factors were speculated to play a role in treatment refractory cases.

Immune responses and chronic Giardia infection

The host defences against *Giardia* infection involve both immunological and non-immunological mucosal processes [24], and disease variability might be partly due to host immune status, which also influences infection susceptibility and clinical severity [25]. Experiments in mice have indicated crucial roles for immunoglobulin A (IgA) and an immunoglobulin transport protein, polymeric Ig receptor, in the host defence against *Giardia* [26]. However, we know little about the function of these and other defence mechanisms, and how they could contribute to, or influence, persistent *Giardia* infection.

Repeated or prolonged exposure seems to produce some protection; children living in endemic areas are more susceptible to disease than are adults, and residents in such areas tend to exhibit lower disease incidence than non-immune visitors [27]. Immunocompromised hosts, such as patients with hypogammaglobulinemia [28], have been associated with a predisposition towards chronic giardiasis, although HIV patients do not seem to show enhanced vulnerability to *Giardia* infection [29]. In a single-patient investigation of treatment-refractory, persistent giardiasis, findings regarding enterotoxicity, immune responsiveness and parasite drug sensitivity were normal, but the capacity of mononuclear cells to kill *Giardia* trophozoites was reduced [30].

A significantly lower serum IgG and IgA has been reported in Indian children with acute and persistent giardiasis, whereas asymptomatic carriers had levels comparable with those of healthy controls [31]. Interestingly, these data also show that persistent cases, despite appropriate chemotherapy, have lower concentrations of *Giardia* membrane protein specific antibodies than acute and asymptomatic cases. Thus, poor ability to produce specific anti-*Giardia* immunoglobulins is a risk factor for persistent giardiasis.

In 40 patients identified with persistent giardiasis after the Bergen epidemic, only one had non-measurable IgA [18]. Thus, there is good reason to believe that predisposition for persistent giardiasis is based not on a single mechanism or deficiency, but rather on a combination of several minor deficiencies of varying degrees in each individual's anti-*Giardia* defences.

Management of treatment-refractory giardiasis

Recurrence of symptoms after treatment could be due to treatment failure, re-infection or syndromes such as post-infectious irritable bowel syndrome (PI-IBS) [32]. If treatment failure is confirmed by a *Giardia*-positive stool sample more than one week after treatment completion, then drug resistance should be assumed (although re-infection should also be considered, particularly in endemic areas), and use of a different class of drug or combination treatment should be considered. It can also be useful to offer a repeat course of the same treatment, for an extended period or at a higher dose.

Clinical and *in vitro* resistance has been documented for all drug classes commonly used for treating giardiasis [5,33]. Although there have been no large, randomised drug trials of treatment-refractory giardiasis, smaller investigations report effective treatment with drugs of different classes or combination therapies. In the Bergen giardiasis outbreak, an observational study of a treatment ladder was conducted on 38 metronidazole-refractory cases [18]. Of these, the majority (79%) responded to albendazole and metronidazole combination therapy. Of those that did not, 50% (3/6) responded to paromomycin therapy, and the remaining three responded to quinacrine and metronidazole combination therapy [18]. In another study of metronidazole-refractory cases, randomization of 20 individuals to either albendazole monotherapy or combination albendazole and metronidazole therapy, demonstrated significantly greater efficacy with the combination [34]. Evidence of an additive effect for metronidazole and quinacrine has also been reported from *in vitro* studies [30].

Refractory giardiasis can be more difficult to treat in immunosuppressed individuals. In a retrospective study of six treatment-refractory cases, four of whom were immunosuppressed, five responded to quinacrine combined with metronidazole or tinidazole [35]. Metronidazole/secnidazole and albendazole-resistant giardiasis in an HIV patient was successfully treated with nitazoxanide [36], and drug resistance of the *Giardia* strain involved was confirmed by *in vivo* and *in vitro* studies. However, *in vitro* resistance might not always correlate with clinical treatment response, and *in vitro* sensitivity testing is generally not available. Thus, based on clinical and *in vitro* evidence of both synergistic effects and cross-resistance between anti-*Giardia* drugs, a combination of drugs from different classes should be used in treatment-refractory cases [33].

Giardia genotype and symptom spectrum

The development of tools to dissect the molecular biology of different *Giardia* isolates, and the knowledge of the spectrum of symptoms associated with giardiasis, has led to the hunt for associations between particular genotypes and defined symptom patterns. The current assimilation of results is inconclusive, with both assemblages associated with diarrhoeal disease. Different symptom spectra are apparently associated with different genotypes in different populations (Table 2). However, in regions with an endemic genotype, a new genotype might cause particularly severe symptoms when it first appears in the population, and dual infection with two different genotypes might produce a synergistic increase in pathology and symptomatology. In addition, the degree of intra-assemblage variation could be of importance, particularly for genotype B *Giardia* [1]. However, even when clear trends can be detected, there are usually exceptions and, thus, these data reinforce again that both host and parasite variables must be included when attempting to understand the interaction between them.

Symptom continuation after successful treatment

Successful giardiasis treatment, with elimination of the parasite from the patient, does not necessarily mean an end to symptoms. Results from a follow-up study after the

Table 2. Associations between symptoms and *Giardia* genotypes. A summary of results from various studies

Study location	Study outline	Genotypic associations with symptoms	Refs
Chandigarh, India	*Giardia* from 6 adults with abdominal symptoms (diarrhoea, abdominal pain, loose stools) and 6 with dermatologic problems but no diarrhoea or gastrointestinal complaints. Genotyping at *tpi* gene (PCR-RFLP).	Gastrointestinal symptoms: 4 assemblage A, 2 assemblage B. Dermatological symptoms: 1 assemblage A, 5 assemblage B.	[42]
The Netherlands	*Giardia* from 9 individuals with persistent diarrhoea, diarrhoea at presentation and more severe symptoms, and 9 with intermittent diarrhoea (alternating episodes) and moderate symptoms. Genotyping at *gdh* gene (PCR-RFLP).	Severe symptoms, persistent/actual diarrhoea: 0 assemblage A, 9 assemblage B. Milder symptoms, intermittent diarrhoea: 9 assemblage A, 0 assemblage B.	[43]
Western Australia	*Giardia* from 23 children. Faecal samples categorised as diarrhoeal or normal. No other symptom data collected. Genotyping at SSU rRNA gene (PCR and sequencing).	Diarrhoea: 6 assemblage A, 3 assemblage B. Normal faeces: 1 assemblage A, 13 assemblage B.	[44]
Turkey	*Giardia* from 20 individuals with diarrhoea and 24 with other conditions (including gastric ulcers {7}, duodenal ulcers {4}, ulcerative colitis {3} and hypertension {3}), but no diarrhoea. Genotyping at *tpi* gene (PCR-RFLP).	Diarrhoea: 17 assemblage A, 3 assemblage B. Other diagnoses, no diarrhoea: 2 assemblage A, 22 assemblage B.	[45]
Northern Portugal	*Giardia* from 7 children with asymptomatic infection. Genotyping at β-giardin gene (PCR and sequencing).	2 assemblage A, 5 assemblage B.	[46]
Dhaka, Bangladesh	*Giardia* mono-infections from 211 individuals, with or without diarrhoea. Genotyping at *tpi* gene (PCR-RFLP).	Diarrhoea: 8 assemblage A, 32 assemblage B. Without diarrhoea: 6 assemblage A, 165 assemblage B.	[47]
Ethiopia	*Giardia* from 43 individuals with abdominal symptoms and 14 asymptomatic. Genotyping at β-giardin gene (PCR-RFLP, some sequencing).	Abdominal symptoms: 19 assemblage A, 12 assemblage B, 3 assemblage F, 6 mixed assemblage A and B, 3 mixed assemblage A and F Asymptomatic: 12 assemblage A, 1 mixed assemblage A and B, 1 mixed assemblage A and F.	[48]
Spain	*Giardia* from 57 individuals with abdominal symptoms and 51 asymptomatic. Genotyping at *tpi* gene (PCR-RFLP).	Abdominal symptoms: 29 assemblage AII, 26 assemblage B. Asymptomatic: 14 assemblage AII, 35 assemblage B. Correlation significant in patients under 5 years.	[49]
Sancti Spiritus, Cuba	*Giardia* from 14 children with abdominal symptoms and 5 asymptomatic. Genotyping at the β-giardin and *gdh* genes (PCR and sequencing).	Abdominal symptoms: 4 assemblage A, 10 assemblage B. Asymptomatic: 4 assemblage A, 1 assemblage B.	[50]
Fortaleza, Brazil	*Giardia* from 41 children. Multiplex real-time genotyping at 18S rRNA gene.	3 Assemblage A infections, 10 assemblage B infections, 5 mixed infections. No correlation with symptoms (presence or duration of diarrhoea).	[51]
Vellore, India	*Giardia* from 50 children with diarrhoea, and 51 asymptomatic. Genotyping at *tpi* gene (PCR-RFLP).	Diarrhoea: 5 assemblage A, 40 assemblage B, 5 mixed infections. Asymptomatic: 2 assemblage A, 48 assemblage B, 1 mixed infection.	[52]

Abbreviations: PCR-RFLP, Polymerase chain reaction followed by restriction fragment length polymorphism analysis; SSU rRNA, small subunit ribosomal RNA.

Bergen outbreak suggest that post-elimination symptom continuation could be more frequent in those who have experienced chronic or treatment-refractory giardiasis [37], presumably because the host physiology has experienced a more prolonged and sustained impact from the parasitosis.

Abdominal symptoms

After the Bergen giardiasis outbreak, at least 124 people (or >9.5% of those diagnosed, and ~5% of all those infected) were referred for medical attention because of a continuation of abdominal symptoms 2–16 months after the acute illness phase [23]. Clinical evaluation of 82 of these people 14–29 months after the outbreak showed continuing abdominal symptoms, particularly diarrhoea-predominant IBS [8]. More surprisingly, two years after the outbreak, 38% of 1017 respondents to a questionnaire reported continuing abdominal symptoms following the *Giardia* infection [38]. Symptoms could not be explained by chronic infection, because all referred cases had eradicated the parasites as previously reported [18]. A similar clinical picture is sometimes observed following amoebic or bacterial gastrointestinal infection [32]. The basis for such post-elimination symptoms requires further investigation in order for the physician to provide appropriate advice or treatment for the patient. Some possible reasons are discussed in Table 3.

Table 3. Possible reasons for symptom continuation after giardiasis treatment

Possible reason	Proposed mechanism	Investigations and their conclusions
Chronic infection	Treatment failure due to insufficient dose or duration of treatment, antimicrobial resistance of parasite or frequent re-infection.	See text (section on chronic giardiasis) and Table 1.
Chronic, cryptic giardiasis	Mixed infections: trophozoites refractory to treatment persist and cause symptoms. Low parasite proliferation (possibly associated with parasite genotype) reduces detection.	Small, randomized open trial investigating effect of treatment on *Giardia*-negative, symptomatic cases after Bergen outbreak. Only temporary symptom relief obtained, presumably due to treatment anti-inflammatory effects; chronic cryptic giardiasis excluded [53].
Mucosal damage and changes in architecture	Giardiasis could result in damage to intestinal epithelial brush border, villous flattening or atrophy and mucosal inflammation. Symptoms could persist after parasite eradication due to prolonged alterations in mucosal tissues. Increased intestinal permeability during infection exposes host to a variety of intestinal antigens and could lead to later hypersensitivity [9].	Experimental studies [21,54]: CD8 T-cell infiltration with cytokine involvement. Parasite attachment to epithelium leads to disruption of tight junctions. *Giardia*-strain dependent epithelial apoptosis. Eosinophilic infiltration, hypercellularity and enterocyte desquamation induced by *Giardia*-antigens demonstrated *in vitro*, possibly promoted by cysteine proteases [54]. Associations between histological findings and symptoms during giardiasis vary in clinical studies [23,54]. In Bergen outbreak cohort, prolonged mucosal inflammation observed in *Giardia*-negative cases with abdominal symptoms [23], and worsening of these symptoms associated with intake of particular food items [8].
Temporary lactase deficiency	Failure to split disaccharides, particularly milk lactose [55] due to epithelial brush border injury and disaccharidase deficiencies. Might cause malabsorption of glucose, sodium and water, with ensuing diarrhoea.	Temporary lactose intolerance commonly seen after giardiasis, possibly mediated by $CD8^+$ T cells [56]. Abnormalities in microvilli brush borders not seen in hosts devoid of functional T cells, even in presence of live parasites, thus intestinal malfunction not merely from parasite virulence factors or trophozoite attachment [57].
Post-infectious irritable syndrome (PI-IBS)	Troubling disorder with recurrent episodes of abdominal pain or discomfort; changes in stool frequency and/or consistency disproportionate to observed pathology. Risk factors include female gender, mucosal lesion severity and past and present psychosocial factors. Increasing evidence for associations with low-grade mucosal inflammation, increased gut permeability and altered serotonin levels [58].	Following the Bergen outbreak, at least 66 patients with persistent abdominal symptoms following successful treatment and parasite elimination fulfilled Rome II criteria for IBS [8]. Repeated duodenal biopsies in 11 patients showed normalisation of initially frequent histological inflammation [8]. Pattern of IBS-subtypes revealed high frequency of diarrhoeal symptoms and little constipation, similar to previously described subtype pattern from patients with PI-IBS following bacterial gastroenteritis [59]. US-based epidemiological study failed to detect IBS following giardiasis, but study design enabled detection of only very severe IBS cases [60].
Bacterial overgrowth	Bacterial overgrowth associated with current and previous giardiasis long postulated as cause for prolonged malabsorption syndrome, 'tropical sprue'.	Intestinal colonisation with enterobacteria found in prolonged cases of giardiasis with malabsorption [61]. Lactulose breath tests in patients with persistent symptoms following Bergen outbreak indicated bacterial overgrowth not responsible for the prolonged symptoms [62].

Fatigue symptoms

Fatigue is commonly reported in giardiasis, but often overlooked. A cluster of cases of chronic fatigue syndrome were speculated to have been precipitated by giardiasis [39], but prolonged fatigue after parasite clearance has only recently been described [38]. In addition, chronic fatigue frequently occurs as a comorbid symptom with IBS, and abdominal complaints are commonly reported from patients with chronic fatigue syndrome. Of 1017 respondents to a questionnaire 2 years after the Bergen outbreak, up to 41% reported fatigue, and abdominal symptoms were strongly associated with fatigue [38]. This association is of interest as there are some common risk factors for development of these complications. Hypochondriasis, adverse life events and depression have all been reported as risk factors for IBS [32]. However, such psychological risk factors seem to be less important in PI-IBS than for development of chronic fatigue syndrome [40]. Although the mechanisms behind fatigue development are obscure, elevations in colonic mast cells in patients with IBS have been correlated with psychological symptoms, including depression and fatigue [41] and deserve further investigation in giardiasis.

Concluding remarks

Giardiasis is one of the most common non-viral causes of diarrhoea, afflicting millions of individuals, worldwide [2]. Although effective treatments are available, the parasite in some cases is refractory to treatment; in addition, debilitating symptoms can sometimes continue even after the parasite has been eliminated, impairing performance and affecting quality of life. In developing countries, this is another burden for an already disadvantaged population.

Although still often considered an uncomplicated parasite, reproducing by simple binary fission, the complexity of *Giardia*, and its relationship with its hosts, is becoming increasingly apparent. As we explore the relationships between genotype and phenotypic characteristics further,

our understanding of the spectrum of pathogenicity in different host populations will increase, and we will be able to address questions on whether postinfectious syndromes (as seen in the Bergen outbreak) are uniquely associated with particular parasite strains, or whether other factors are important, and, if so, which (for Outstanding Questions, see Box 1). Host characteristics, including immune status, underlying medical conditions, previous infection with other *Giardia* strains and/or other intestinal pathogens, and other factors are almost certainly also of importance. One mechanism postulated for the PI-IBS following the Bergen outbreak, is that the immune activation occurring during the initial infection was not completely resolved and a low-grade inflammation ensued. Usually only invasive microbes (*Salmonella, Campylobacter, Shigella)* result in PI-IBS, and we might postulate that the normally non-invasive protozoan *Giardia* only results in PI-IBS, and accompanying fatigue, if there is an inflammatory host reaction initially.

That ~40% of people infected in the Bergen outbreak report fatigue and IBS-like symptoms two years after successful treatment is remarkable; presumably a lack of follow-up studies has meant this has not previously been noted in other outbreaks in developed countries. It is possible that the sequelae known from developing countries, such as the impacts on growth and cognitive function in children, have simply seemed irrelevant to the Western world, and therefore similar later effects of infection have not previously been investigated. Outbreaks of giardiasis occur infrequently in developed countries, but sporadic cases are frequent. Thus the widespread occurrence of giardiasis in developing countries means that data from such areas can provide insights of value to developed, as well as developing, countries, although the differences in patient populations must not be overlooked.

Box 1. Outstanding questions

Although our understanding of the spectrum of symptoms associated with giardiasis has improved markedly in recent years, there are still vast gaps in our knowledge. We believe that future studies should address the following:

- Treatment options: how often does treatment failure occur in sporadic cases and how should this best be addressed?
- How and to what extent do the phenotypic/genotypic characteristics of *Giardia* isolates alter in a population over time?
- How does the pathogenicity of different *Giardia* isolates vary?
- How can new biomolecular methods be best applied to investigate this variation?
- Is there a synergistic interaction between different *Giardia* isolates, such that infection with two different isolates results in considerably greater pathogenicity?
- What are the patient risk factors for chronic infection and for postinfection sequelae?
- How are the differences in host immune responses to *Giardia* infection influenced by the following variables: patient age (children/adults), previous exposure (naïve/experienced populations), *Giardia* genotype and immunogenic *Giardia* proteins?
- What is the extent and severity of chronic fatigue syndrome and IBS in giardiasis compared with these conditions when associated with other aetiologies?
- What are the similarities or differences between postinfectious learning disabilities in children in developing countries and post-giardiasis chronic fatigue syndrome?
- What are the most important triggers for excystation and establishment of infection? Why do some *Giardia* genotypes establish in some hosts and not in others?

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