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CASE REPORT

Blastocystis sp. subtype 2 cause of diarrhea in patient referred to GI clinic

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ABSTRACT

Blastocystis hominis is a protozoan parasite mainly found in human fecal samples both from symptomatic patients and asymptomatic population in developing countries. In this study we report a patient referred to GI clinic with abdominal pain, nausea, vomiting, fever for 2 or 3 days and passed watery stools because of Blastocystis sp. subtype 2 infection.

Key words: Blastocystis sp, genotype, diarrhea, IBD

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Introduction:

Blastocystis hominis is accepted as the aetiological agent of gastrointestinal symptoms when it is found in high numbers in the stool and no other agent presented. It is a common microscopic parasite found in human faecal samples, both symptomatic patients and healthy people throughout the world. If all pathogens are ruled out, it will be possible to link it with human disease (1).

According to molecular analysis, at least ten subtypes (STs) were classified for Blastocystis hominis (2). However, a clear-cut linkage of distinct STs to the presence of disease symptoms is not apparent (3). More recently, intra-subtypes variability with pathogenic and nonpathogenic variants within certain STs (4) as well as intra-subtypes difference in morphotypes (5) has been suggested to be related to the pathogenic potential of Blastocystis. In this study we report a patient referred to GI clinic with abdominal pain, nausea, vomiting, fever for 2 or 3 days and passed watery stools because of Blastocystis sp. subtype 2 infection.

Case Report:

A 65-year-old Iranian woman was admitted to gastroenterology unit located in the Taleghani hospital which was referred from a rural health center with possible infection with IBD.

The patient was complaining for abdominal pain, nausea, vomiting, fever for 2 or 3 days and at last passed watery stools. On physical and laboratory examination, there was right lower quadrant tenderness and rebound. Her body temperature was 38.58C, white blood cell count was 16.200/mm^3, C-reactive protein was 77 mg/L. She underwent endoscopy and colonoscopy any cause of diarrhea including IBD and celiac disease, but all her findings including endoscopy, colonoscopy and histological examination were normal. At the next step, a microbiological examination was performed on three consecutive stool samples including bacterial enteropathogens, enteric viruses (Astro-, Adeno-, Rota- and Norovirus), Shiga and Clostridium difficile toxins, helminthes and intestinal protozoans (Entamoeba histolytica, Dientamoeba fragilis, Giardia intestinalis, microsporidia, and cryptosporidia). For parasitological evaluations all three times samples were examined using direct wet mount for presence of B.hominis and formalin-ether concentration method for helminthes eggs and protozoan cysts. By bright field microscopy B.hominis was detected at ×400 magnification, applying the criterion of five or more organisms per high-power field as diagnostic index. No other established or potential bacterial, viral, or parasitic pathogens or any toxins were detected. For cultivation, positive samples were cultured at 37 c in Hsr+ s (horse-serum-Ringer- starch) medium, as modified by Diamond (1993), with no egg albumin. After 48h, the parasites were harvested by centrifugation (at 300×g for 5 min), washed twice with saline, re-suspended in distilled water, and stored at 4c until the DNA extractions (6). After culturing B.hominis proliferation, DNA was extracted using phenol/chloroform/isoamyl alchol and precipitated with ethanol /sodium acetate prior to PCR reaction (6). Each DNA sample was run in a standard PCR with the spesific primers. The thermocycler used was set for 35 cycles each of 1 min (3 min in the first cycle) at 93°C, 1 min at 55°C and 2 min at 72°C. The products were separated by electrophoresis on 1.2% agarose gels. The PCR products were then sequenced using an Applied Biosystems (ABI) Terminator Cycle Sequencing Ready Reaction kit (BigDye® Terminator V3.1 Cycle Sequencing Kit) on an ABI 3130xl Genetic Analyzer. The sequences obtained were manually edited and aligned using gunrunner software. The sequencing study confirmed the B.hominis subtype 2 and main cause of diarrhea in this patient. Treatment with metronidazol and co-trimoxazole was initiated and her symptoms significantly improved.

Discussion:

B. hominis has vague role in human disease. It may be usually detected in stools during a workup for gastrointestinal symptoms. The B. hominis clinical presentations principally include abdominal pain and diarrhea. Also atypical GI symptoms such as nausea, anorexia, vomiting, weight loss, lassitude,

dizziness, and flatulence can be seen in these patients. Different studies have suggested a pathogenic role of B. hominis in causing intestinal inflammation. Also some reports have suggested there is an association between B. hominis and inflammatory bowel disease (IBD) and/or irritable bowel syndrome (IBS) (7).

The role of B. hominis in human intestinal disease is controversial. Several clinical and epidemiological studies implicate B. hominis as a pathogen while others dismiss it as a commensal (1-5). However, the literature remains anecdotal and most of the studies were uncontrolled. Nagler et al. examined the hospital course of 12 patients with exacerbated IBD, who also had stool specimens positive for B. hominis to determine the effect of B. hominis on their disease. All patients responded suitably to medical therapy but corticosteroid treatment did not worsen the condition in any case. Their results showed that B.hominis is not a significant pathogen in IBD and treatment must be directed toward the underlying illness (8).

Irritable bowel syndrome (IBS) is a functional gastrointestinal disorder in which abdominal pain is associated with a defect or a change in bowel habits. Gut inflammation is one of the proposed mechanisms of pathogenesis. Recent studies have described a possible role for protozoan parasites, such as B. hominis and Dientamoeba fragilis, in the etiology of IBS.

Immune activation was demonstrated by Hussain et al. (1997) who showed that IgG antibody levels to B. hominis in patients with IBS were significantly elevated compared with asymptomatic controls (9). Endoscopic biopsies have demonstrated edema and inflammation in the colon and small bowel based on IBS presentation (10).

On the other hand, recent clinical studies have reported the association of skin rash with Blastocystis infection (11).

The transmission, pathogenicity, culture characteristics, taxonomy, life cycle, biochemistry and molecular biology of B. hominis remain unclear. According to these findings we suggest that the stools of all patients presenting with IBD or IBS should be examined, and genetic study should be carried out for B. hominis. Therefore, more extensive case-controlled studies are required to clearly define the potential pathogenicity of B. hominis in intestinal disease and IBS.

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