Gastrointestinal Basidiobolomycosis, a Rare and Under-diagnosed Fungal Infection in Immunocompetent Hosts: A Review Article

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Abstract
Gastrointestinal Basidiobolomycosis (GIB) is an unusual, rare, but emerging fungal infection in the stomach, small intestine, colon, and liver. It has been rarely reported in the English literature and most of the reported cases have been from US, Saudi Arabia, Kuwait, and Iran. In the last five years, 17 cases have been reported from one or two provinces in Iran, and it seems that it has been undiagnosed or probably unnoticed in other parts of the country.

In this review, we explored the English literature from 1964 through 2013 via PubMed, Google, and Google scholar using the following search keywords:

1) Basidiobolomycosis
2) Basidiobolus ranarum
3) Gastrointestinal Basidiobolomycosis

In this review, we attempted to collect all clinical, pathological, and radiological findings of the presenting patients; complemented with previous experiences regarding the treatment and prognosis of the GIB.

Since 1964, only 71 cases have been reported, which will be fully described in terms of clinical presentations, methods of diagnosis and treatment as well as prognosis and follow up.

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Keywords
● Gastrointestinal ● Entomophthorales ● Basidiobolus ranarum ● Immunocompetent

Introduction
Basidiobolomycosis is a rare fungal infection caused by Basidiobolus ranarum.1

The Zygomycetes includes two fungal orders: Mucorales and Entomophthorales, with completely different pathogenic potentials. Mucorales involve only the immunocompromised patient, while Entomophthorales, which include Basidiobolus genera, causes infection in immune competent individuals, mostly chronic infection of the subcutaneous tissue.2-4 This fungal infection has been extremely rare in immunocompromised hosts such as diabetics and renal transplant patients.5 Just the opposite, Mucormycosis has been rarely reported in immunocompetent hosts with no underlying disease.6-8

Basidiobolus ranarum (B. haptosporus, B. meristoporus) is a fungus belonging to the order Entomophthorales in the family of Zygomycota.5,10 Basidiobolus species are types of filamentous

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fungi belonging to the family of Basidiobolaceae of the order Entomophthorales. Basidiobolus ranarum was first described in 1886 in frogs. This fungus is an environmental saprophyte found in soil and decaying vegetable materials. This fungus is endemic in some parts of the world such as India, China, and Indonesia. It is commonly found in soil and decaying vegetable materials and occasionally found as a commensal in the gastrointestinal tracts of amphibians, reptiles, fish, dogs, frogs, and bats. Even fatal cases have been reported in toads. B. ranarum is proven as a low virulent fungus. It has been identified in mice, i.e. after inoculation of this fungus in experimental mice; invasive disease has not occurred.

The first recognized human case of infection caused by Basidiobolus ranarum was reported from Indonesia as a subcutaneous infection in 1956. Since then, there have been few individual case reports regarding subcutaneous, nose, and sinus infection by this opportunistic fungus. Typically, this fungus causes chronic subcutaneous infection mainly in children as young as one month of age. Without treatment, the subcutaneous infection could be similar to soft tissue tumors; and thus, prompt treatment of the skin is essential. Even huge subcutaneous infections have been reported to cause perineal intestinal obstruction or intraperitoneal spread. Additionally, sometimes delayed treatment can cause disfigured extremities.

It seems that, this fungal infection is acquired by exposure to B. ranarum after minor trauma to the skin or by biting insects. This rare fungus can also rarely infect other parts of the body especially gastrointestinal tract.

In 1964, the first documented case of gastrointestinal Basidiobolomycosis (GIB) was reported in a 4-year-old boy. Thereafter, it was progressively identified and the number of case reports increased from different parts of the world such as Saudi Arabia, Kuwait, USA, and Iran. However, no definite and characteristic clinical presentation has been reported and all previous cases have been operated without initial diagnosis of GIB. Preoperative diagnosis has been different neoplastic and nonneoplastic diseases such as colorectal cancer and Crohn's disease, inflammatory pseudotumor, etc.

In this review, we extensively gathered information from previously reported cases in the literature with the emphasis on the demographic findings, presenting symptoms, laboratory findings, diagnostic tests, and treatment options. We conducted a comprehensive search of medical literature from 1964 through May 2014 via PubMed, Google, and Google scholar using the following search keywords:

1. Basidiobolomycosis
2. Basidiobolus ranarum
3. Gastrointestinal Basidiobolomycosis

All cases with the diagnosis of gastrointestinal Basidiobolomycosis in immunocompetent host were included in the review. Cases published more than once (duplicate case reports) were excluded. All important findings such as presenting symptoms, method of primary diagnosis, laboratory findings, operative findings, treatment modalities, and follow up studies were separately included in each case.

Since the first report of GIB in 1964 until the present, there have been 71 cases of GIB, in the English literature. Cases have been reported from Saudi Arabia with 23 cases, Iran with 17 cases, USA with 23 cases, Kuwait with 2 cases, The Netherlands with one case, and Iraq with 6 cases. The patients were ranged from 1.5 to 80 years old. This disease was significantly more common in males; from 71 reported cases, only six patients were female.

Nearly all patients had abdominal pain and fever as their initial symptoms; however, some cases additionally had constipation (4 cases), diarrhea (1 case), and gastrointestinal bleeding (1 case). Only one patient was presented with mucoid stool. Furthermore, one case was presented with perforated appendicitis because of fungal invasion in the appendiceal wall.

Radiologic examination by ultrasonography and CT scan were reported in 20 cases as intestinal wall thickening in 8 and intestinal, gastric or abdominal masses in other 12 cases. There were 3 cases with preliminary diagnosis of inflammatory bowel disease such as Crohn's disease with and without fistula. In 12 reported cases, there were concomitant liver and intestinal masses.

Most cases had preoperative endoscopy and biopsy as diagnostic procedures, none of which led to any diagnoses. There were 3 cases with incorrect preliminary diagnosis of tuberculosis in the colon biopsy because of the presence of granuloma in the pathologic examination. The erythrocyte sedimentation rate (ESR) was 33 to 138, and eosinophil count was <1 to 27.7%. It is worthy to note that only in 5 cases eosinophil count was normal.

The method of diagnosis was pathology and culture. In 32 cases, culture was performed, however only 50% turned out to be positive. Pathologic diagnosis of the cases was characteristic and showed the same picture in all of them, i.e. the presence of Splendore-Hoepli bodies and many eosinophils as well as intensely radiating eosinophilic granular
material surrounding the fungal elements. This histological finding is very characteristic of this fungus and other invasive fungi with gastrointestinal involvement such as mucormycosis causes extensive necrosis and vascular invasion and even their granulomas are morphologically different with no eosinophile.

Recently, there are reports of PCR (polymerase chain reaction) and molecular diagnosis of GIB, with high specific and sensitivity. Also, there is a report of ultrastructural features of GIB. On ultrastructural level, fungal hyphae, spores, and macrophage-laden crystalloids could be seen.

An extremely rare report on successful experimental diagnosis of this infection by immunodiffusion is published, but so far, no commercial test is available for immunologic diagnosis of B. ranarum infection.

There are reports of elevated levels of cytokine responses such as IL4, IL10, TNFα (TH2 type cytokines) after infection with B. ranarum that can be helpful as a diagnostic test.

Most of the cases (59 patients) undergone surgery with intestinal, and gastric or liver resection, and all cases received antifungal therapies including itroconazole and amphotericine B, but rarely ketoconazole, pescoconazole and fluconazole. There are a few reports on the successful treatment with Polymixin E.

Among these 71 patients, eight cases deceased, probably due to delayed treatment and because of the disseminated disease. The deceased cases were pediatric except for one who was 61 years old. Five of the previously reported patients had recurrence, despite of fungal medical therapy and surgery. Table 1 shows the most common findings in the previously reported cases of GIB.

**Table 1: Most common characteristics of gastrointestinal Basidiobolomycosis in previously reported cases**

| Reported findings                  | Most common characteristic       |
|-----------------------------------|----------------------------------|
| Presenting symptoms               | Abdominal pain and fever         |
| Age                               | 1.5-80 years                     |
| Sex (M/F)                         | 65/6                             |
| ESR                               | 33-138                           |
| Eosinophils                       | 1-27.7%                          |
| Most common preliminary diagnosis | Intestinal, gastric or liver mass|
| Culture positive cases            | <50%                             |
| Pathologic diagnosis before surgery by endoscopy | 0
| Characteristic histological findings | 100%                           |
| Recurrence                        | 4 cases                          |
| Died                              | 8 cases                          |

Discussion

There are more than 100,000 fungi worldwide, only 150 of which are pathogen. Basidiobolomyosis is a rare fungal infection due to B. ranarum, an environmental saprophyte. It is a member of the order Entomophthorales of the class Zygomycetes. B. ranarum is a well-described fungus in the skin and subcutaneous tissue; however, this fungal infection in the gastrointestinal tract is an emerging infection.

It seems that most reported cases have been from parts of the world with warm climate, such as Arizona in the US, Fars province in Iran, Saudi Arabia, and Kuwait. We believe that this is due to the nature of the fungus, which mostly lives in a warm and humid environment and involves human GI tract by ingestion of contaminated fruits, vegetables, and water.

Another important concern is the route of acquiring this infection. The proposed theories are ingestion of infected food (e.g. people with pica) and using contaminated papers for cleaning the skin (e.g. toilet papers). Some of the previous studies reported the infection years after a surgical procedure. Therefore, implanted fungus after surgery is another less likely theory. Another proposed theory is the history of ranitidine ingestion, which can decrease gastric acidity and allow fungal survival after gastric passing. In addition, Smoking can decrease mucosal WBC function and facilitate fungal infection by B. ranarum. Previous reports from Arizona (USA) were on people working as gardener and landscaper. Consequently, work environment is another unproven exposure.

Since the first case of GIB, about 71 cases have been reported most of which are during the last 10 years. It seems that preoperative diagnosis of GIB by radiologic and clinical features is relatively impossible. Most common imaging findings on this disease have been concentric intestinal, gastric wall thickening or polypoid mass where all are nonspecific findings. The most important point that causes delayed diagnosis in GIB is its occurrence in immunocompetent host. This fungus involves immunocompetent and healthy hosts and all previously reported cases were on patients in good health except for a few
reports on diabetic patients.\textsuperscript{31} Conversely, most of fungal infections in GI tracts have been reported in the immunosuppressive patients. Thus, it is very difficult for the clinicians to think about a fungal infection in the GI tract of a completely healthy patient.\textsuperscript{50} \textit{B. ranarum} lies deep beneath the mucosa, so endoscopic biopsies are non-representative, and in none of the previously reported cases, the endoscopic biopsy was diagnostic.\textsuperscript{34}

Morphology of \textit{B. ranarum} shows hyphae and zygospores. Hyphae shows branching and septation with thin wall. The whole hyphae are surrounded with an eosinophilic and amorphous hyalinized material. Zygospores have foamy cytoplasm with a large nucleolus.\textsuperscript{32}

Although histological features of this infection has been well characterized in the skin and subcutaneous tissue, but its presence in the GI tract is challenging for pathologists. Even, there were cases with the erroneous biopsy diagnosis of tuberculosis, simply due to the presence of granulomatous reaction the mucosa.\textsuperscript{33,34} In our unpublished experience, even in cases with preoperative biopsy, erroneous diagnoses of ameba, tuberculosis and even Crohn's disease is very common.

The main histological findings of the pathologic sections stained with hematoxylin and eosin stain in gastrointestinal Basidiobolomycosis are as below:

1) Marked infiltration of eosinopills
2) Mixed infiltration of PMN leukocytes and granulomatous inflammation
3) Thin wall and broad hyphae surrounded by eosinophilic material, which are easily seen by hematoxylin and eosin staining, however Periodic Acid-Schiff (PAS) and Gomori Methenamine Silver (GMS) can intensify the fungal wall staining\textsuperscript{43}
4) Zygospores which are very similar to trophozoites of amoebae\textsuperscript{32}

This fungal infection has been misdiagnosed and underdiagnosed; the reason for such delayed diagnosis is multifactorial:

1) Nonspecific clinical manifestation
2) No definite risk factor
3) Negative endoscopy and colonoscopy (submucosal growth)\textsuperscript{37}

The most common and best diagnostic clue for the diagnosis of this fungal infection is primarily by considering this disease in differential diagnosis. However, common presenting clinical and paraclinical findings in nearly all previously reported cases were the same; that is abdominal pain and fever in a patient with abdominal, gastrointestinal or colon mass or intestinal wall thickening who has concomitant significantly high ESR and eosinophilia.\textsuperscript{62}

The gold standard for definite diagnosis of GIB is culture.\textsuperscript{41,42} For best results of the culture, the tissue should be inoculated to the culture media very soon, as it cannot survive at 4°C.\textsuperscript{60} After 2 to 3 days of incubation in sabouraud agar at 20 to 30°C, white to pale grey colonies with radial folds would appear.\textsuperscript{31}

However, in most previous experiences, the patient has undergone surgery with the preoperative diagnosis of mass, cancer, or inflammatory bowel disease. Therefore, the specimen has been sent to a lab in formalin and no culture has been performed, however, even in cases with culture, negative results were common.\textsuperscript{1,11,12}

The pathologic findings after surgery are characteristic, and in combination with clinical and laboratory tests can be diagnostic. Consequently, in the cases with no available culture or negative culture histopathologic findings can aid to avoid treatment delay.\textsuperscript{40}

The only fungal infections, which are somehow similar in histology to \textit{B. ranarum}, are \textit{Conidiobolus coronatus}, \textit{Conidiobolus incongruous}, and \textit{Pythium insidiobolus}. These fungi are seen in the head and neck only in immunocompromised hosts.\textsuperscript{31} Viseral involvement of these fungi has not been reported.\textsuperscript{45}

Immunologic diagnosis of \textit{B. ranarum} with methods such as immunodiffusion has been reported, which seems to be specific and has no cross reactivity with other fungi of Entomophthorales; however, its sensitivity is controversial. This immunodiagnostic test can also be helpful for follow up of patients.\textsuperscript{43}

Molecular diagnosis has also been performed in some reports with optimum results by DNA extracted from formalin fixed paraffin embedded tissue, however, due to the rarity of the disease; many centers do not have the set up for this method.\textsuperscript{63}

Another important concern with this fungal infection is prompt treatment by combined surgery and medical therapy to eradicate disease and prevent early recurrence. Delayed treatment can cause disseminated disease, which has life-threatening outcome and causes postmortem diagnosis after autopsy studies.\textsuperscript{54} Additionally, delayed treatment can cause complications such as bowel perforation, obstructive uropathy, esophageal varices, and duodenobiliary fistula.\textsuperscript{1}

It seems that overall diagnosis of fungal infections is increasing even in immunocompetent hosts. Fortunately, there has been an increase in the number of broad-spectrum antifungal agents allowing for better therapeutic choices.\textsuperscript{65-67}

On this specific fungus (i.e. Basidiobolus ranarum), there have been diverse experiences
about the choice of antifungal therapy. Even rare reports on medicinal plants indicate its success as a form of treatment. Very few studies have reported successful treatment with amphotericin B, though with resistance in more than 50% of the treated cases. It seems that the best regimens involve pre- and postoperative itroconazole for at least 6 months. Prolonged treatment can prevent recurrence, but early discontinuation of the antifungal therapy is reported to cause recurrence of the disease even after surgery. Until now, no fungal resistance has been reported by itroconazole. Recently, another new antifungal with successful treatment with posaconazole is reported.

**Conclusion**

In this review, gastrointestinal Basidiobolomycosis is introduced as an emerging infection, and its life threatening and aggressive nature is emphasized. It requires prompt diagnosis and surgery; otherwise, it can be fatal with delayed and inadequate treatment. In addition, there are unresolved questions in this fungal infection, the most important of which is predisposing factors for its acquisition in healthy people. This question needs more experience and diagnosing more cases to find out the exact predisposing factors.

**Conflict of Interest:** None declared.

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