Gastrointestinal basidiobolomycosis in a patient suffering from duodenal ulcer with perforation: First case report from Italy

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INTRODUCTION

Gastrointestinal basidiobolomycosis (GIB), an unusual fungal infection caused by the fungus Basidiobolus ranarum, is rarely reported in the medical literature. GIB is difficult to diagnose because its clinical presentation is non-specific and has no identifiable risk factors. We report here the first case of GIB diagnosed in Italy in a patient suffering from a duodenal ulcer with perforation. The patient was successfully treated with itraconazole. The absence of non-specific signs and symptoms of GIB may delay a definitive diagnosis and treatment. A microbiological investigation should always be requested in order to reach a rapid and definitive diagnosis and to rule out other intestinal diseases.

Table 1 - Main characteristics of the 122 GIB cases reported in the literature from 1964 to 2018.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>No. Patients (Proportion %)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Symptoms</td>
<td></td>
</tr>
<tr>
<td>Abdominal pain</td>
<td>110 (90%)</td>
</tr>
<tr>
<td>Abdominal mass</td>
<td>49 (40%)</td>
</tr>
<tr>
<td>Laboratory tests</td>
<td></td>
</tr>
<tr>
<td>Culture total</td>
<td>82 (67%)</td>
</tr>
<tr>
<td>Positive culture</td>
<td>47 (57%)</td>
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<tr>
<td>Histopathology total</td>
<td>116 (95%)</td>
</tr>
<tr>
<td>Positive Histopathology</td>
<td>115 (99%)</td>
</tr>
<tr>
<td>Therapy and outcome</td>
<td></td>
</tr>
<tr>
<td>Azole</td>
<td>110 (90%)</td>
</tr>
<tr>
<td>Death</td>
<td>20 (16%)</td>
</tr>
</tbody>
</table>

Key words: Fungal infection; Basidiobolomycosis; Duodenal ulcer; Basidiobolus ranarum.
below the first case of GIB diagnosed in Italy in an immuno-
nocompetent man suffering from a duodenal ulcer with perforation.

**CASE REPORT**

A 78-year-old Caucasian Italian man presented to the Accident and Emergency department of our hospital for abdominal pain, vomiting and loss of appetite. Medical history revealed a 30-year history of abdominal pain episodes caused by a gastric ulcer treated with antacids (aluminum hydroxide plus magnesium hydroxide and rabeprazole sodium) and prostatic hypertrophy treated with finasteride. Initial laboratory tests were normal (blood count, prothrombin activity, serum electrolytes, serum creatinine, biochemical liver and pancreas function tests) except for elevated serum procalcitonin (PCT) levels (4.72 ng/ml). After surgery, laboratory tests were still normal and PCT levels decreased at 48 h (1.60 ng/ml) and 72 h (0.93 ng/ml). An abdominal x-ray obtained with the patient in a supine position revealed evidence of a pneumoperitoneum with gas under the subdiaphragmatic region. The findings were suggestive of a duodenal ulcer with perforation. The patient was handed over to the surgical team, who administered ceftriaxone (2 gr. i.v.) and paracetamol (1000 mg i.v.) therapy. The diagnosis of peritonitis was based on laparoscopic findings of purulent fluid in the peritoneal cavity, and a sample of peritoneal fluid was sent to the microbiology laboratory in two BacT/ALERT blood culture bottles for aerobic and anaerobic germs (bioMérieux, Marcy-l’Étoile, France). The blood culture bottles were incubated in a BacT/ALERT VIRTUO (bioMérieux, Marcy-l’Étoile, France) detection system. The aerobic bottle, which positivized after 34 hours, was cultured on blood agar (B), chocolate agar (C), MacConkey agar (M) and Sabouraud’s dextrose agar (S). The B, C, and M culture media were incubated at 37°C and the S at 30°C. The colonies grew rapidly on B, C and S agar, attaining a diameter of 2-3 mm within 18 hours; their color was greyish and opaque, rugose with a waxy texture (Figure 1). No growth was observed on M agar. As the growth progressed, the formation of many satellite colonies was observed due to forced ejection of sporangioles (balanostromes). Slide cultures revealed a large number of asexual spores (sporangiospores), some of which had a knob-like adhesive tip (Figure 2). Based on these microscopic and macroscopic findings, a zygomycete belonging to the order of Entomophthorales (probably *Basidiobolus spp.*). In cases with a 100% negative predictive value and 65% positive predictive value for sepsis caused by Candida spp. The identification of the isolate was then confirmed by DNA sequencing. DNA amplification and sequencing of the ITS region (internal transcribed spacer) was carried out by the Institute of Microbiology at the Catholic University of the Sacred Heart in Rome. The fungus-specific universal primers ITS1 (5′-TCCGTAGGTGAACCTGCGG-3′) and ITS4 (5′-GCAATATCAATAAGCGGAGGA-3′) were used to amplify the ITS region (Leaw et al., 2006). Amplification was performed using the Hotstart Taq Master Mix Kit (Qiagen, Hilden, Germany). The amplified DNA was sequenced using the ABI PRISM 3130xl Genetic Analyzer (Applied Biosystems, Foster City, California, USA) tool. The sequences were compared to reference data available at the GenBank database using BLAST (http://www.ncbi.nlm.nih.gov/BLAST/). Post-operatively, the patient did well and was successfully treated by targeted therapy with itraconazole (200 mg administered twice per day).

**DISCUSSION**

The presumptive identification of a zygomycete belonging to the Entomophthorales order was performed by microscopy examination just three days after the sample had been received by the microbiology laboratory. The identification of *B. ranarum* by DNA sequencing led to the definitive diagnosis of GIB in a patient who had never traveled out of Italy. In most cases described in the literature, patients had leukocytosis and eosinophilia (Vikram et al., 2012; Geramizadeh et al., 2012; Geramizadeh et al., 2015; Mohammadi et al., 2018). In our case report, the patient’s laboratory tests were normal, except for PCT. Although PCT is not a specific indicator of fungal infections, it can be induced in persistent and invasive fungemia (Gérard et al., 1995; Ortega et al., 2004; Distefano et al., 2004). Charles et al. (2006) found a significantly lower PCT level in patients with candidemia (median 0.65 ng/ml) compared to those with bacteremia (median 9.75 ng/ml). A PCT level higher than 5.5 ng/ml demonstrated a 100% negative predictive value and 65% positive predictive value for sepsis caused by Candida spp. In cases
where PCT is 1-5 ng/ml in conjunction with a fungal infection, its decrease could be related to the success of the therapy (Jemli et al., 2007). In our case, the decrease in PCT levels followed the surgery, as the targeted therapy with itraconazole was started three days after the surgical treatment. However, other cases of patients with severe fungal infections report low or non-existent levels of PCT (Huber et al., 1997; Beaune et al., 1998). The patient described in this case report had 30-year history of abdominal pain episodes and a gastric ulcer treated with antacid. Abdominal pain, the most common presenting symptom of peptic ulcer disease with antacid. Abdominal pain, the most common presenting symptom of peptic ulcer disease with antacid. Abdominal pain, the most common presenting symptom of peptic ulcer disease with antacid. Abdominal pain, the most common presenting symptom of peptic ulcer disease with antacid. Abdominal pain, the most common presenting symptom of peptic ulcer disease with antacid.

The gold standard for definite diagnosis of GIB remains microbial culture of B. ranarum or surgical specimens (Zabolinejad et al., 1996). The patient had no unusual exposure to soil and denied contact with any animal species in which the organism is known to be a saprophytic colonizer. The mode of acquisition of the disease remains poorly understood. To confirm a case of GIB, the key element is either the characteristic histopathological appearance of the biopsy (tissue biopsy shows eosinophilic infiltration and typical Splendore–Hoepli phenomenon) or isolation of B. ranarum from specimens (Vikram et al., 2012). In our case report, GIB was diagnosed only by isolation of B. ranarum from the patient’s peritoneal fluid samples (no histopathological exams were carried out). Bigliazzi et al. (2004) reported the first case of disseminated basidiobolomycosis in Italy. In this case, cultures and serologic tests for B. ranarum or other Entomophthorales agents had not been carried out because of clinical suspicion of neoplasia or vasculitis. The diagnosis was made on the basis of autopsy findings. The gold standard for definite diagnosis of GIB is microbial culture of B. ranarum from fresh aspiration or surgical specimens (Zabolinejad et al., 2014; Al-Maani et al., 2014) and requires a high level of clinical suspicion by the clinician and the mycologist. B. ranarum does not survive at 4°C. Sabouraud agar is an adequate medium, and visible growth is usually present 2-3 days after incubation at 25-30°C (Kwon-Chung et al., 1992). Because of the nonspecific signs and symptoms presenting, GIB can cause intestinal perforation and may masquerade as another clinical entity, delaying definitive diagnosis and treatment. If untreated, the mortality rate of disseminated disease approaches 16%. In order to consider GIB in the differential diagnosis, we suggest microbiological investigation in cases of abdominal pain with a mass (Mortada et al., 2011), and a clinical profile suggestive of malignancy (Nemenqani et al., 2009).

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References


