Impact of Zygomycosis on Microbiology Workload: a Survey Study in Spain

Marta Torres-Narbona, Jesús Guinea,* José Martínez-Alarcón, Patricia Muñoz, Ignacio Gadea, and Emilio Bouza as a representative of the MYCOMED Zygomycosis Study Group

Clinical Microbiology and Infectious Diseases Department, Hospital General Universitario Gregorio Marañón, Universidad Complutense, Madrid, Spain

Received 11 December 2006/Returned for modification 2 March 2007/Accepted 20 March 2007

This multicenter, population-based study evaluated the laboratory workload produced by zygomycetes and the number of cases of zygomycosis in Spain during 2005. Less than 8% of the patients who harbored zygomycete isolates had zygomycosis. The incidence of zygomycosis (6 cases) was 0.43 cases/1,000,000 inhabitants and 0.62 cases/100,000 hospital admissions.

Zygomycosis, a life-threatening infection caused by molds of the class Zygomycetes (2, 17, 22, 24, 28), presents only sporadically (19). More recently, however, a remarkable increase in its incidence has been reported from single institutions in the United States and Europe (9, 10, 12–16, 21, 23, 26, 27). The widespread use of voriconazole for the prophylaxis of invasive fungal infections in those institutions has been claimed to be responsible for this problem (10, 14, 23).

Furthermore, the frequency of isolation of these fungi in clinical microbiology laboratories and the proportion of patients who harbor such isolates and who really have zygomycosis are not reported.

We aimed to clarify the workload produced by zygomycetes in clinical microbiology laboratories to determine their clinical significance and to determine the incidence of zygomycosis in a population-based study in Spain.


Study period and participating hospitals. We invited 140 hospitals to participate in the study, and 50 (36%) participated. The 50 hospitals cared for 14,069,094 inhabitants and were uniformly distributed throughout Spain (which has 44 million inhabitants) (3). The institutions were classified as follows: more than 1,000 beds (9 hospitals), between 500 and 1,000 beds (18 hospitals), and less than 500 beds (23 hospitals). All institutions were general hospitals, and 48 were tertiary-care and/or teaching hospitals. Among the 50 participating hospitals, 28 had bone marrow transplant units. The size and geographical distributions of the nonparticipating institutions and the participating centers were similar.

Information requested. Every hospital completed a preestablished form that allowed us to assess the clinical and microbiological information for each hospital for the whole of 2005. The microbiological data included the number of samples processed during the study period, the number of samples processed for the isolation of fungi, the number of samples with fungal isolates, the number of samples from which zygomycetes were isolated, the number of patients with one or more samples positive for zygomycetes, and the number of samples from patients with zygomycosis. Information regarding the population that the hospital cared for, the number of beds, and the number of total admissions during 2005 was requested. All isolates were derived from clinical samples.

Zygomycetes were identified by conventional methods, based on morphological features.

We requested the completion of a preestablished protocol for every patient meeting the diagnostic criteria of proven or probable invasive fungal disease, according to the EORTC/MSG international standards developed for the diagnosis of invasive aspergillosis in hematological patients (1). In summary, a diagnosis of zygomycosis may be achieved by use of a combination of clinical evidence of the disease and detection of the fungus in tissues (either by histopathological findings or by isolation of zygomycetes from clinical samples).

Clinical and microbiological data were requested for each patient with zygomycosis.

Evaluation of incidence of zygomycosis and description of the clinical forms. During the study period, 78 patients harbored one or more isolates of zygomycetes, but only 6 patients (7.7%; 95% confidence interval [CI], 1.8 to 13.6) from four different institutions had zygomycosis. Five of the six patients had clinical evidence of the infection and the fungus was isolated from clinical samples microbiologically. The other two patients had clinical evidence of infection, and a histopathological diagnosis was made without microbiological isolation (Table 1). This enabled us to calculate an incidence of 0.43 cases per 1,000,000 inhabitants/year (95% CI, 0.00 to 1.72) and 0.62 cases/100,000 hospital admissions.

Overall, five of the six patients had malignant hematological conditions (83%), including leukemia and lymphoma. Five of the patients were under immunosuppressive treatment, three had neutropenia, and only one had diabetes and had received corticosteroid treatment. Only one of the six patients had received voriconazole as prophylaxis during the 15 days before

* Corresponding author. Mailing address: Servicio de Microbiología Clínica y Enfermedades Infecciosas-HIV, Hospital General Universitario Gregorio Marañón, c/ Dr. Esquerdo, 46, 28007 Madrid, Spain. Phone: 0034-914265104. Fax: 0034-913721721. E-mail: jguineaortega@yahoo.es.

Published ahead of print on 28 March 2007.
the diagnosis. Of the six cases, four died and two were cured by surgery plus antifungal treatment.

**Workload evaluation.** The participating hospitals cared for a population of 14,069,094 inhabitants, and their microbiology laboratories processed 2,815,873 samples. Of these, 187,031 (6.6%) were processed for the isolation of fungi. Overall, 37,192 (19.9%) of them were positive for yeasts and/or molds. Of the 50 hospitals, only 19 (38%) reported the clinical isolation of zygomycetes from 171 samples (0.45%). The laboratories isolated 12.2 zygomycetes per 1,000,000 inhabitants (95% CI, 5.36 to 19.06), 6 per 100,000 total samples processed in the microbiology laboratory (95% CI, 2 to 10), 0.62 per 100,000 hospital admissions (95% CI, 0.00 to 1.3), and 5 per 1,000 samples submitted for fungal isolation (95% CI, 1 to 9). Of the 171 positive samples, only 6 were from infected patients and the probability of having zygomycosis when a culture was positive for zygomycetes was 3.5%; only 7.7% of the patients from whom zygomycetes were isolated had zygomycosis.

This population-based study shows that the workload generated by zygomycetes in Spanish clinical microbiology laboratories is very low, and only a small proportion of patients from whom a zygomycete is isolated from one or more clinical samples presented with zygomycosis. The study also shows the low incidence of zygomycosis.

Data that can be used to compare the workload produced by zygomycetes in microbiology laboratories are scarce. Zygomyces can be isolated from the environment. Previous studies showed that they can be isolated in 20% to 39% of outdoor air samples (4), 1.6% to 9.5% of hospital air samples (18), and occasionally as colonizers in human samples (11). The isolation of zygomycetes in clinical laboratories should be interpreted with caution.

Recently, an increased incidence of zygomycetes has been reported in individual institutions or even in individual units within those institutions (6–10, 12–16, 20, 23, 27, 29). The incidence of zygomycosis in the United States was estimated to be about 1.7 cases per 1,000,000 inhabitants between 1992 and 1993, or about 500 patients per year (19). When the disease is diagnosed postmortem, it is 10- to 50-fold less frequent than candidiasis or aspergillosis and is supposed to be present in one to five cases/10,000 autopsies (5, 25, 30).

Institutions reporting on the increased incidence of zygomycosis usually relate it to the widespread use of voriconazole as antifungal prophylaxis (12, 15, 23, 29). The rate of consumption of voriconazole/1,000,000 inhabitants in 2005 was 3,227 daily defined doses in Spain (G. Armenteros, Pfizer Spain, personal communication), while in the United States it was 5,506 daily defined doses. This difference may explain the low incidence of zygomycosis in Spain, but without population-based studies evaluating its incidence in the United States or studies specially designed to clarify this issue, the reason for this difference remains a suggestion.

Our data suggest that the reemergence of zygomycosis cannot be assumed from reports from single units or single institutions. In Spain, voriconazole is not widely used for antifungal prophylaxis. A word of caution regarding the widespread use of voriconazole for prophylaxis seems pertinent.

The staff of the microbiology services of all participating hospitals are members of the MYCOMED Zygomyces Study Group and are as follows: A. Sánchez Porto, Hospital del S.A.S. La Línea de la Concepción, Cádiz; R. M. Gordillo Urbano, Hospital Universitario Reina Sofía (S.A.S.), Córdoba; M. de la Rosa Fraile and C. Miranda Casas, Hospital General Virgen de las Nieves, Granada; I. Cuesta Lendínez and L. Parras Jiménez, Hospital Universitario Ciudad de Jaén, Jaén; V. Pérez Valero and P. Blanc Iribarren, Hospital Regional Carlos Haya, Málaga; A. Pascual Hernández and A. I. Suárez Barrenechea, Hospital Universitario Virgen Macarena, Sevilla; E. Martín Mazuelos and C. Martín Martín de la Escaleria, Hospital Universitario Nuestra Señora De Valme, Dos Hermanas, Sevilla; M. J. Revillón Pinilla and A. Rezusta López, Hospital

---

**Table 1. Clinical and microbiological data for the six patients with true invasive zygomycosis**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Clinical form</strong></td>
<td>Rhinosinusal</td>
<td>Rhinosinusal</td>
<td>Pulmonary and cutaneous</td>
<td>Pulmonary</td>
<td>Cutaneous-facial</td>
<td>Cutaneous-abdominal</td>
</tr>
<tr>
<td><strong>Underlying condition</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Neutropenia</td>
<td>Yes</td>
<td>Yes (chronic lymphatic leukemia)</td>
<td>Yes (acute myeloid leukemia)</td>
<td>Yes (acute myeloid leukemia)</td>
<td>Yes (lymphatic leukemia)</td>
<td>Yes (non-Hodgkin’s lymphoma)</td>
</tr>
<tr>
<td>Hematological malignancies</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Transplant</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Diabetes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Imaging data (X ray, CT scan, NMR)</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Previous immunosuppressive treatment</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Previous antifungal treatment (30 days previously)</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Previous antibiotic treatment (30 days previously)</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Previous corticosteroid treatment</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Samples</td>
<td>Rhinosinusal sample</td>
<td>Not done</td>
<td>None</td>
<td>Wound</td>
<td>Sputum (r = 3)</td>
<td>None</td>
</tr>
<tr>
<td>Histopathology</td>
<td>Not done</td>
<td>Presence of hyphae</td>
<td>Not done</td>
<td>Not done</td>
<td>Not done</td>
<td>Not done</td>
</tr>
<tr>
<td>Diagnostic method</td>
<td>Clinical and culture</td>
<td>Clinical and histopathology</td>
<td>Clinical and culture</td>
<td>Clinical and histopathology</td>
<td>Clinical and culture</td>
<td>Clinical, culture and histopathology</td>
</tr>
<tr>
<td>Outcome</td>
<td>Cured</td>
<td>Cured</td>
<td>Cured</td>
<td>Cured</td>
<td>Cured</td>
<td>Cured</td>
</tr>
</tbody>
</table>

---

* a BMT, bone marrow transplant.

b CT, computed tomography; NMR, nuclear magnetic resonance.
This study does not present any conflict of interest for the authors.

We thank Thomas O’Boyle and Laurence Baron for help with the preparation of this manuscript.

This study was partially financed by grant C/03/14 from Red Espanola de Salud, Madrid, Spain.

This work was supported by the National Institutes of Health grants AI45470, AI45471, AI45472, AI45473, and HL55550, the Veterans Affairs Health Services Research and Development Service, and the Leland H. Stromberg Memorial Fund.

Reports of the National Institute of Neurological Disorders and Stroke Study Group and the Multicenter Trial of L-1-ValSubmitted on October 15, 2017 by guest http://jcm.asm.org/ Downloaded from

REFERENCES


