

Best Practice Recommendations for Medical Mycology

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Description

Zygomycosis is an important emerging fungal infection, associated with high morbidity and mortality. The Working Group on Zygomycosis of the European Confederation of Medical Mycology (ECMM) prospectively collected cases of proven and probable zygomycosis in 13 European countries occurring between 2005 and 2007. Cases were recorded by a standardized case report form, entered into an electronic database and analysed descriptively and by logistic regression analysis. During the study period, 230 cases fulfilled pre-set criteria for eligibility.

Haematological Malignancies

The median age of the patients was 50 years; 60% were men. Underlying conditions included haematological malignancies (44%), trauma (15%), haematopoietic stem cell transplantation (9%) and diabetes mellitus (9%). The most common manifestations of zygomycosis were pulmonary (30%), rhinocerebral (27%), soft tissue (26%) and disseminated disease (15%). Diagnosis was made by both histology and culture in 108 cases (44%). Total mortality in the entire cohort was 47%. On multivariate analysis, factors associated with survival were trauma as an underlying condition, treatment with amphotericin B and surgery; factors associated with death were higher age and the administration of caspofungin prior to diagnosis. In conclusion, zygomycosis remains a highly lethal disease. Administration of amphotericin B and surgery, where feasible, significantly improve survival.

For the classification of each case as proven or probable, the revised definitions of invasive fungal disease of the European Organization for Research and Treatment of Cancer/Mycosis Study Group (EORTC/MSG) were used, with the following modification: If the diagnosis was made by histology and there was PCR testing on tissue positive for zygomycetes (from the central laboratory described above), the case was classified as proven zygomycosis, even if there were no cultures available. In addition, diabetes mellitus was also included in the host criteria.

Diagnosis by histology was made if large, non-septate hyphae were reported. The sites of infection were classified as recently described by Roden et al. Mortality was assessed as all-cause mortality during the course of zygomycosis.

Statistical Analysis

Statistical analyses were conducted using spss. Differences between the qualitative variables in two or more groups were analysed by a chi-square test. A two-sided p value of <0.05 was considered significant. For the estimation of predictors of outcome the following method was used. Univariate analysis was performed using logistic regression for each variable separately. Variables entered into univariate analysis included age, sex, underlying diseases, treatment with corticosteroids, immune suppressives or antifungals prior to the diagnosis of zygomycosis, sites of infection and types of treatment. The variables for which a statistically significant relationship was shown (p<0.05) were used to construct a new multivariate model using the logistic regression approach. The multivariate results presented are those from the most parsimonious model, including only the selected covariates.

In each participating European country, a national coordinator was appointed, who prospectively collected zygomycosis cases, recorded by the treating physicians in standardized Case Report Forms (CRFs), which were then sent either by email or fax to the general study coordinator. The national coordinators were all experts in the field of zygomycosis and in most cases were appointed by the respective national Mycology Societies. The participating hospitals were selected by the national coordinator on the basis of their capacity to document all episodes of zygomycosis occurring during the study period.

Mucorales isolates were initially identified at the participating institutions by the routine methods used in each laboratory. Molecular identification was then performed either at a national centre or the isolates were sent to a central laboratory in Spain for sequencing. Furthermore, coordinators were informed that they could send paraffin-embedded tissue for PCR identification to a central laboratory in Germany.

In order to be included as a case in the registry, sufficient information regarding diagnosis, predisposing factors and clinical presentation had to be provided. The CRFs were reviewed by the principal investigators and queries were sent to the national coordinators in order to complete missing data. After completing the database, a data review committee examined all the data. The study was approved by the ethics committee of the University of Athens 'Laikon general hospital', in Athens, Greece, the institution of the principal investigators. In addition,

approval was also obtained from local ethics committees of all collaborating countries according to local regulations.

Diabetes was the sole predisposing factor in 21 (9%) patients. In another 18 (8%), diabetes was combined with other underlying conditions, such as malignancy or trauma. Of the 39 cases with trauma, 18 had surgical trauma and 4 had various underlying diseases (non-Hodgkin lymphoma, Ewing sarcoma, solid organ transplantation and diabetes). The remaining 16 patients were immunocompetent and trauma was the sole predisposing factor for zygomycosis. In another two patients no predisposing factor was found. Thus, in total, there were 18 (8%) patients that were considered immunocompetent.

Invasive fungal diseases are an important cause of morbidity and mortality in a wide range of patients, and early diagnosis and management are a challenge. We therefore did a review of the scientific literature to generate a series of key

recommendations for the appropriate use of microbiological, histological, and radiological diagnostic methods for diagnosis of invasive fungal diseases. The recommendations emphasise the role of microscopy in rapid diagnosis and identification of clinically significant isolates to species level, and the need for susceptibility testing of all *Aspergillus* spp, if treatment is to be given. In this Review, we provide information to improve understanding of the importance of antigen detection for cryptococcal disease and invasive aspergillosis, the use of molecular diagnostics for aspergillosis, and the crucial role of antibody detection for chronic and allergic aspergillosis. Furthermore, we consider the importance of histopathology reporting with a panel of special stains, and emphasize the need for urgent (<48 hours) and optimised imaging for patients with suspected invasive fungal infection. All 43 recommendations are auditable and should be used to ensure best diagnostic practice and improved outcomes for patients.