Susceptibility testing of *Aspergillus niger* strains isolated from poultry to antifungal drugs – a comparative study of the disk diffusion, broth microdilution (M 38-A) and Etest® methods

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Abstract

The aim of this study was to determine the sensitivity of *Aspergillus niger* strains isolated from birds to available antifungal drugs using different *in vitro* assays – classical disk diffusion, Etest® and broth microdilution NCCLS/CLSI M 38-A. The study material consisted of about 2,000 swabs and samples from different species of birds. *A. niger* (n=10) was accounted for 6.81% of the total pool of strains isolated. Determinations were made for 13 antifungal drugs using the disk diffusion method. The *A. niger* exhibited high susceptibility to enilconazole, terbinafine, voriconazole, tioconazole and ketoconazole, low susceptibility to clotrimazole, miconazole and nystatin, and resistance to amphotericin B, itraconazole, pimaricin, fluconazole and 5-fluorocytosine. Minimum inhibitory concentration (MIC) was determined for 9 antifungal drugs using the micromethod of duplicate serial dilutions in a liquid medium. *A. niger* strains were most susceptible to enilconazole and voriconazole. MIC ranged from 0.0625 to 0.5 μg/ml for enilconazole, with MIC<sub>90</sub>0.5 μg/ml and MIC<sub>50</sub>0.125 μg/ml. The corresponding values for voriconazole were 0.25-1 μg/ml, 1 μg/ml and 0.5 μg/ml. MIC for amphotericin B and terbinafine ranged from 0.5 to 4 μg/ml, while the values for the remaining drugs were highly varied. MIC was measured by the gradient diffusion method using Etest® for 5 antifungal drugs: amphotericin B, fluconazole, itraconazole, ketoconazole and voriconazole. By far the highest susceptibility was obtained in the case of voriconazole, with MIC ranging from 0.0625 to 1 μg/ml. MIC for amphotericin B ranged from 0.25 to 4 μg/ml, for itraconazole and ketoconazole ranging from 0.5 to 16 μg/ml. Methods available for this purpose are not always applicable in field conditions. The present results indicate that the Etest® technique, due to its high percentage of agreement with the M 38-A microdilution method, should find application in medical and veterinary practice.

Key words: *Aspergillus niger*, poultry, disk diffusion method, broth microdilution (M 38-A) method, Etest®

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