Infrared Spectroscopy for Fast Characterization of Fungi Important in Hygiene and Agricultural Sciences Using Micro-techniques

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Fungi play an important role within our environment. The mycelium of some species produces extracellular enzymes that are capable to decompose even degradation-resisting polymers and substances such as lignin, cellulose, various other carbohydrates, oil products, pesticides and others. Fungi are also known for their involvement in various biochemical processes. On the other hand, fungal pathogens such as *Fusarium spp.* can cause severe diseases to various plants resulting in serious damage within a large number of crops and are responsible for infections in humans or can be allergenic. Concerning hospital-acquired infections, invasive fungal infections resulting from filamentous fungi, such as *Aspergillus spp.*, remain serious infections despite recent therapeutic progress. The risk of acquiring such infections is especially relevant to immuno-compromised patients.

Conventional species identification is time-consuming and subjective, since it requires culturing for several days and is based on the subjective assessment of morphological characters. Culturing the pathogens on selective nutrient media, and morphology examination of the fungal colony using an optical microscope is among the current classical methods used for the detection and identification of fungi.

In contrast, Fourier transform infrared spectroscopy (FTIR) was used here to identify and differentiate important fungal pathogens. IR-spectroscopy based methods are rapid by using reagent-free nondestructive techniques, which require minimal technical training and sample preparation. The analysis is usually not labour-extensive in comparison to existing techniques that require advanced technical competence.¹⁻³

FTIR measurements were performed either in the attenuated total reflection (ATR) mode using a silverhalide-fiber coupled diamond micro-prism in combination with dry-film preparation for ensuring good contact of the mycelium to the ATR-element or using the potassium bromide micro-pellet method with small amounts of the dry fungus mycelium ($\sim 100~\mu g$) that was pestled with about 10 mg of KBr powder. Using these methods, the infrared bands assigned to proteins, lipids and carbohydrates varied in wavenumber position, intensity and line shape among the species studied. Since growth medium, cultivation method, growth stage or strain degeneration may strongly influence spectral variability and experimental reproducibility, so first of all, a standardized sample preparation was developed. Based on such a protocol, spectral results using both measurement techniques will be illustrated and discrimination by cluster analysis will be reported.

References

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