

# **Industrial and Food Microbiology and Biotechnology**

### O-34 - AGED FREEZE-DRIED AMPOULES OF PRESERVED BIOTECHNOLOGICAL IMPORTANT FUNGI

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## Background

The implementation of consistent fungal preservation techniques and appropriate quality assurance are key issues for an effective and efficient preservation. The cost and convenience of each method are important aspects to be taken into consideration such as the knowledge of all parameters capable of affecting the procedures [1]. Preservation methods currently used are highly empirical and in many instances, do not provide reliable genetic and phenotypic stability. Freeze-drying is commonly used to preserve fungal strains at room temperature, however, genetic and phenotypic alterations after long term-storage are yet unknown. Therefore, the main goal of the present experimental study is to evaluate the freeze-drying preservation method for the effective long-term preservation of strains belonging to *Aspergillus* section *Nigri*.

#### Method

Twenty-one strains representative of *Aspergillus* section *Nigri* were selected and preserved by freeze-drying. The strains were subjected to accelerated storage during 4 weeks at 37 °C. These samples were morphological, physiological and genotypical analysed. In order to detect macro and micro-morphological changes, growth for seven days at 25°C on Potato Dextrose Agar, Malt Extract Agar, Czapek Yeast Extract Agar and Czapek Dox Agar was performed. The physiological changes were monitored for the detection of ochratoxin A and fumonisin B2 as described elsewhere [2,3]. In order to identify genotypic changes, DNA fingerprinting techniques using the oligonucleotides M13 and (GACA)<sub>4</sub> were performed. All assays were evaluated at 3 points in time: before preservation (I), 2 (II) and 4 (III) weeks after preservation.

### **Results & Conclusions**

For all the methodologies used to evaluate freeze-drying of fungi along time the major results are: 1) no significant changes were observed in the macro and micro-morphological analysis; 2) all strains maintained their mycotoxins production pattern, before and after ageing; 3) after ageing different DNA fingerprinting was observed.

In conclusion, freeze-drying can be considered a technique of excellence to be used on the maintenance of biodiversity within the filamentous fungi, and more accurately for *Aspergillus* section *Nigri*. However, it is recommended to consider possible genetic changes after long shelf-life periods.

### **References & Acknowledgments**

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