Dynamics and Biodiversity of microorganisms (fungi, yeast and bacteria) by PCR-DGGE, influencing OTA production on coffee beans

Durand Noël¹, El Sheikha Aly^{1,2}, Fontana Tachon Angélique¹, Montet Didier¹

¹ UMR Qualisud (CIRAD, Montpellier University II), 34095 Montpellier Cedex 5, France.

32511 Shibin El Kom, Egypt.

Résumé

Ochratoxin A is a secondary metabolite produced by various filamentous fungi contaminating in a wide range of food and animal feedstuffs. OTA has been shown to possess nephrotoxic, carcinogenic, immunodepressive and teratogenic properties (1).

In tropical zones, OTA is mainly produced in coffee beans by three Aspergillus species: *A. carbonarius*, *A. niger* section Nigri and *A. ochraceus* section Circumdati. Among them, the most important OTA producer and the most frequently isolated is *A. carbonarius*. In temperate zones *Penicillium verrucosum* and *P. nordicum* are known to synthetize OTA in food commodities(2, 3).

The OTA content in coffee was shown to be closely link to harvesting conditions, postharvest processing conditions and especially dry processing, storage and transportation conditions (4, 5, 6). In some producing countries, damaged caused on beans by other fungal communities undoubtedly lead to high OTA contents in coffee.

In order to understand the OTA contamination process in foodstuffs, PCR-DGGE (Polymerase Chain Reaction - Denaturing Gradient Gel Electrophoresis) assays were carried out on coffee microflora. PCR-DGGE is a rapid molecular technique that was developed to monitor the dynamics of microbial populations (fungi, yeast and bacteria). PCR-DGGE is used to characterize the microbial flora of food products by extraction and amplification of 16S, 26S and 28S rDNA for bacteria, yeast and fungi. PCR-DGGE stages i.e. extraction and amplification were optimized. Detection limits are estimated on several fungus, yeast, and bacteria. Additionally, repeatability and sensibility of the methodology were also tested. In this study only fungi methodology are showed.

² Department of Food Science and Technology (Minufiya University, Faculty of Agriculture),