



## THE ISOLATION OF *ASPERGILLUS* SPP FROM HARVESTED MAIZE IN THREE PORTUGUESE REGIONS

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### KEYWORDS

Maize, *Aspergillus* species, Mycotoxins.

### INTRODUCTION

In Portugal, maize constitutes one of the most important crop and it is the cereal that involves more agriculture explorations from the North to the South of the country.

Maize is colonized and contaminated by different species of fungus including *Aspergillus*, *Fusarium*, and *Penicillium* (Gioni *et al.* 2007). *Aspergillus* spp., among other species, are usually associated with this cereal, during its drying and storage. The most important *Aspergillus* spp. are *Aspergillus flavus* and *Aspergillus parasiticus*, due to the fact that, together with *Aspergillus nomius*, are the main producers of aflatoxins, a mycotoxin proven to have toxic effects in mammals (Sapamundo *et al.* 2007). It is also known that *A. flavus* species can produce cyclopiazonic acid, a mycotoxin that occurs naturally in maize and peanuts as a co-contaminant with aflatoxins (Vaamonde *et al.* 2003).

To prevent the fungi's development and the subsequent production of mycotoxins it is important to monitor the production chain and to create a predictive model, based in two of the most important abiotic factors, water activity and temperature. The aim of this project is to study the level of contamination of maize with aflatoxigenic fungi right after harvesting and during storage time, in three Portuguese regions, allowing with this approach, the prevention of occurrence of mycotoxins in the final product.

### MATERIALS AND METHODS

The survey was carried out in three Portuguese regions, with producers belonging to the National Producers Association of Maize and Sorghum (ANPROMIS –

Associação Nacional de Produtores de Milho e de Sorgo):

- A - Cooperativa Agrícola de Coimbra (Coimbra, Coimbra district)
- B - AGROMAIS (Riachos/Torres Novas, Santarém district)
- C - CERSUL (Santa Eulália/Elvas, Portalegre district)

The samples were collected between November 2008 and April 2009.

### RESULTS AND DISCUSSION

- The sampling occurred in three distinct places of the storage chain
- Maize samples were plated in MEA10 (malt extract medium agar with 10% of NaCl) and the resulting fungi were isolated to MEA;
- Presence of mycotoxins was tested by high profile liquid chromatography (HPLC), by growing the *Aspergillus* spp. isolates in YES (for aflatoxins) and CYA (for OTA and CPA)
- It was possible to obtain, from a total of 132 samples, 1075 isolates from the gender *Aspergillus* and 732 of this isolates were tested for mycotoxins;
- The isolates were divided in three distinct groups: *Aspergillus* section *Flavi* (tested for CPA and aflatoxins), *Aspergillus* section *Nigri* (tested for OTA) and Other *Aspergillus* (tested for CPA and OTA).

□ Results show that there are differences between the incidence of the three groups of *Aspergillus* in the regions. Whereas in Elvas (Portalegre) and Riachos (Santarém) there is a high incidence of *Aspergillus* section *Nigri*, the same doesn't happens in Coimbra. This may be explained by the fact that black *aspergilli* are more resistant to the higher solar exposure and higher temperatures, typical of these regions. *Aspergillus* section *Flavi*, are more common in the samples after leaving the dryers. Most of them are producers, the great majority of CPA and less than 50% of aflatoxins. A very small percentage of *Aspergillus* section *Nigri* are OTA producers even though they are abundant in the samples of two regions; *Aspergillus* section *Flavi* are very common in all regions even though they are more common after drying and storage. The great majority of fungi are not producers, but there is an alarming quantity of producers of aflatoxins and CPA.

## CONCLUSION

It is possible to correlate the climate with the kind of isolates obtained, being *Aspergillus* section *Nigri* associated with the regions of hotter and dryer climates.

## REFERENCES

Richard, E., Heutte, N., Bouchart, V., Garon, D., Evaluation of fungal contamination and mycotoxin production in maize silage, *Animal Feed Science and Technology* 148 (2009) 309–320;

Sapamundo, S., Devlieghere, F., Geeraerd, A., Meulenaer, B., Impe, J. F., Debevere, J., Modelling of the individual and combined effects of water activity and temperature on the radial growth of *Aspergillus flavus* and *A. parasiticus* on corn, *Food Microbiology* 24 (2007) 517-529

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