CHARACTERIZATION AND DYNAMICS OF FUSARIOSIS ON MAIZE IN BELGIUM

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The BCCM™/MUCL collection

This research program is realized at the Mycothèque de l'Université catholique de Louvain (MUCL), that is part of the Belgian Co-ordinated Collections of Micro-organisms (BCCM™) * consortium. The MUCL collection contains more than 28,000 living strains of filamentous fungi and yeasts of agro-industrial or ecological interest. Standard and control strains used in bioassays, deterioration and biodegradation are also available.

Introduction

Fusariosis is one of the most important problems encountered in maize crops in Belgium and it is responsible for significant losses (up to 30%) in both grain yield and quality. More than 7% of the world cultures are destroyed by fusariosis (Hoffmann and Schmutterer 1999 ; Papst *et al.*, 2005).

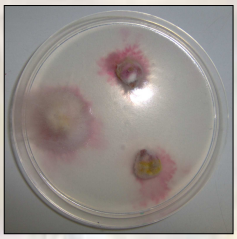
Fusariosis is in fact the result of simultaneous contaminations by several *Fusarium* species, mostly *F. graminearum*, *F. verticillioides* and *F. proliferatum*. Other species are also detected at low frequencies, such as *F. avenaceum*, *F. subglutinans*, *F. sambucinum*, *F. sporotrichioides*, *F. crookwellense*, *F. culmorum*, *F. poae*.

Furthermore, recent studies suggest that a wider diversity than that already known could exist within some species or species complex. For example, *F. graminearum* has been recently subdivided in nine phylogenetic species (O'Donnell *et al.*, 2004) ; the *Gibberella fujikuroi* species complex (containing *F. verticillioides*) was subdivided in at least 46 phylogenetically species from which 10 sexual species (Leslie and Summerell, 2006).

Some of those *Fusarium* species are able to produce mycotoxins, potentially responsible of human and animal health diseases. Important amounts of those mycotoxins are often found in maize silages, causing health problem for the livestock. Nevertheless, it is established that the physico-chemical properties existing in silages (pH, aerobiose, t °) prevent the growth of *Fusarium* spp. (Scudamore and Livesey, on 1998). Mycotoxins are thus produced in the field.

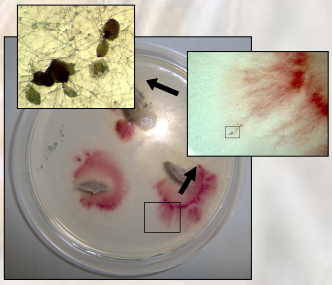
The present project aims to understand the dynamics of the various *Fusarium* species development during the cultural season in Belgium, the genetic variability within species, as well as their mycotoxin production. Two partners are associated with BCCM™/MUCL to the present project supported by the Région Wallonne **: The CARAH (Centre Agronomique de Recherches Appliquées du Hainaut) and the CIPF (Centre Indépendant de Promotion Fourragère, UCL) who are in charge of field assays.The CARAH team also performs the mycotoxin detection and quantification.

Materials and methods



An extensive assay has been performed in Belgium in five different eco-climatic situations. Eight maize varieties presenting a range of susceptibility to fusariosis have been collected at four different physiological stages (flowering, ~26% dry matter content, ~33% beginning harvest, ~40% end of harvest), for detection and identification of the *Fusarium* species, and for quantification by HPLC (and possible correlation) of the four main *Fusarium* mycotoxins : zearalenone, deoxynivalenol, fumonisins and T2-toxin.

Small fragments of the stalk, ear and silks from each maize plant were surface sterilized and put on Petri dishes for isolation of the fungal flora. Pure *Fusarium* strains were preserved on PDA and SNA in glass tubes, under sterile mineral oil.

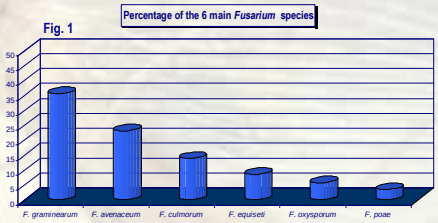


Results and discussion

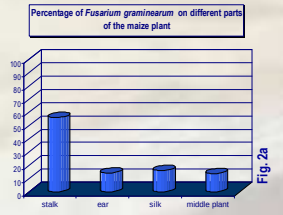
A total of approximately 2200 *Fusarium* spp. strains have been isolated and are preserved at BCCM™/MUCL. Preliminary fungal identification and mycotoxin quantification results are available for the flowering stage.

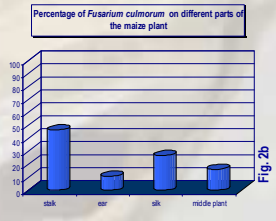
On basis of a national fusariosis evaluation, the year 2005 has been quoted as particularly favourable for the development of *Fusarium* spp., what is correlated with the observation of this very **early and unusual installation** of the pathogens on plants. Indeed, an extremely high number of plants (55% to 95%) were contaminated by *Fusarium* spp., while no symptoms were visible.

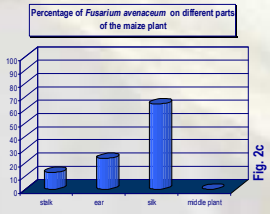
Furthermore, a **wide spectrum of *Fusarium* species** has already been identified **(Fig. 1)**. *F. graminearum*, known as the most important pathogen on maize, represents 35,5% of the identified strains. *F. avenaceum*, which is responsible of the "*Fusarium* head blight" on wheat, represents 23,1% of the *Fusarium* strains. Important percentages of *F. culmorum* (13,9%), *F. equiseti* (8,6%) and *F. poae* (3,5%) were also detected. Other species were occasionally detected (<2%) : *F. redolens, F. arthrosporioides, F. venenatum, F. torulosum, F. crookwellense, F. ramigenum. F. oxysporum* which is a pathogen of many plants and crops but which is not known as an important threat for maize, reaches 5% of the identified *Fusarium* strains.

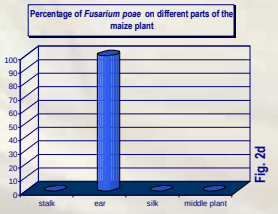


The percentage of contaminated plant bases is higher than that of ear contamination, suggesting an extension of the *Fusarium* spp. from the base to the top of the plant. Moreover, the different species of *Fusarium* tend to **develop on different parts of the plant**. Indeed, *F. graminearum*, *F. equiseti* and *F. culmorum* develop mainly on maize stalks **(Fig. 2a, 2b)**, *F. poae* and *F. avenaceum* are almost exclusively observed on ear and silks **(Fig. 2c, 2d)**.









Preliminary **mycotoxin analyses** in plant organs from which the *Fusarium* spp. strains have been isolated do not allow detecting zearalenone, although various producing species were present.

Conclusion

An unsuspected level of *Fusarium* contamination was observed on maize plants from the early stage of crops. Together with the observation that a very important number of species has been detected, it raises the question of a possible underestimated pathogenicity and mycotoxin threat caused by *Fusarium* in fields.

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