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ABSTRACT

In the frame of a research project aiming at detection and control of Fusarium and related mycotoxins in banana fruits and maize in China, plant samples were collected in four provinces. In the southern provinces of Hainan and Yunnan, cultivated and wild banana plants coexist, together with maize. In Hubei and Henan, the Fusarium diversity was investigated on maize at the end of the growing season. Beside 14 Fusarium species, an important number of isolates belonging to the Fusarium incarnatum-equiseti complex species (FIESC) was detected. In order to understand the impact of this finding at both the genetic and mycotoxigenic levels, their phylogeny was studied on basis of the Elongation factor-1a (EF-1a) gene, and the AFLP technique was used to provide a preliminary picture of their genetic diversity on maize.

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Genetic and mycotoxigenic diversity of isolates belonging to the *Fusarium incarnatum-equiseti* species complex, and recovered from maize and banana in China

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Introduction

In the frame of a research project aiming at detection and control of *Fusarium* and related mycotoxins in banana fruits and maize in China, plant samples were collected in four provinces (Fig. 1). In the southern provinces of Hainan and Yunnan, cultivated and wild banana plants coexist, together with maize. In Hubei and Henan, the *Fusarium* diversity was investigated on maize at the end of the growing season.

Beside 14 *Fusarium* species, an important number of isolates belonging to the *Fusarium incarnatum-equiseti* complex species (FIESC) was detected. In order to understand the impact of this finding at both the genetic and mycotoxigenic levels, their phylogeny was studied on basis of the Elongation factor-1e (EF-1e) gene, and the AFLP technique was used to provide a preliminary picture of their genetic diversity on maize.

Results

Besides numerous other fungal species, a total of 204 *Fusarium* isolates belonging to 15 species were obtained from the four visited provinces. Interestingly, the taxonomic status of 83 isolates remains uncertain. Indeed, their EF-1e were highly similar (>99%) or even identical to isolates of the *Fusarium incarnatum-equiseti* species complex from human beings, investigated recently by O’Donnell et al. (2009), or from sorghum (Funnel-Harris et al., 2010).

An important phylogenetic diversity was observed (Fig. 2): 33 EF1-haplotypes were distinguished, grouped into 8 clusters (I to VIII), from which 7 were already described as phylogenetic species (MLST) by either O’Donnell et al. (2009) or Funnel-Harris et al. (2010). One new phylogenetic species (VI), represented by 5 isolates was detected on banana. Most isolates belong to the EF1 clade while only 1 isolate belongs to the equiseti clade.

No host specificity was observed: several strains from banana and maize belong to the same phylogenetic species, since some others belong to different ones. There was no correlation with geographical origin.

For maize, the 4 phylogenetic species found were perfectly supported by the AFLP clustering (Fig. 3). A similarity of less than 60% confirms a true species rank of these isolates. Furthermore, an important diversity was observed within groups, without any correlation with the geographical origin of the isolate origin.

Using PCR, it was demonstrated that the strains isolates s of the FIESC groups have NO FUM gene.

Material & Method

- **Identification**: the monocausal *Fusarium* spp. isolates were molecularly identified using the protocols described in EF-1e gene sequences.
- **Phylogeny**: restriction analyses (PAUP 4.0b10) were performed separately for isolates from the 3 plant hosts (F. equiseti, F. culmorum and F. incarnatum) into the analysis; F. culmorum was used as outgroup (O’Donnell et al., 2004). See legend Fig. 2 for details of the analysis.
- **AFLP**: after restriction with Eco RI and Mse I, ligations, pre-selective amplification, selective amplification (Eco RI-Mse I: 450-B500; Bgl II-Mse I: 600-900) and capillary gel electrophoresis, the polymeric bands, visualized by Gelography I.4.6, were scored 1/0. See legend Fig.3 for details of the UPMA genetic analysis.
- **Mycotoxic potential**: PCR detection of the *FUMZ* gene was performed using two home-made primer genes targeting the *FUMZ* gene in the fusarium gene cluster: 
  >FUMZ1tac, 5’-TTTATCAGCTCATTTACACG-3’; >FUMZ1six, 5’-AAGCTGTCATCTACGAC-3’

Discussion and perspectives

The molecular results obtained on a large number of isolates allow to conclude that the MLST previously described could be reconsidered as species. AFLP data analyses are in progress for the banana isolates, and will be compared to those on maize. The similarity of isolates from human beings and plants should require more investigations!

Members of the FIESC have been reported in literature to produce type A and B trichothecene mycotoxins (O’Donnell et al., 2009) which is of great concern about their possible origin of the trichothecenes detected (or not) in maize in China, besides those produced by *F. graminearum*. The occurrence of trichothecenes on maize is also in question.

The occurrence of FIESC isolates on both banana and maize, often cultivated near to each other, raises the question of cross contamination banana<->maize, and of the epidemiology of the disease.

Finally, the occurrence of *Fusarium* on wild banana, in an area of origin of the host, questions the occurrence of a possible coevolution phenomenon between both organisms.

References