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Identification of aflatoxigenic and non-aflatoxigenic strains of *Aspergillus* Section *Flavi* isolated from Portuguese almonds

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Abstract

Aspergillus subgenus *Circumdati* section *Flavi*, also referred to as the *A. flavus* group, has attracted worldwide attention for its industrial use and toxigenic potential. Section *Flavi* is divided in two groups of species. One includes the aflatoxigenic species *A. flavus*, *A. parasiticus* and *A. nomius*, which cause serious problems in agricultural commodities, and the other one includes the non-aflatoxigenic species *A. oryzae*, *A. sojae* and *A. tamarii*, traditionally used for production of fermented foods. Differentiating aflatoxigenic from non-aflatoxigenic species and strains in food commodities is of major importance in food quality control. A polyphasic approach consisting of morphological, chemical and molecular characterization was applied to 31 isolates of *Aspergillus* Section *Flavi* originating from Portuguese almonds, with the aim of characterizing and identifying aflatoxigenic and non-aflatoxigenic strains. On the basis of morphological characters, we found two distinct groups among the population under study: 58% were classified as *A. parasiticus* and the remaining 42% were classified as *A. flavus*. Chemical characterization involved the screening of the isolates for aflatoxins B (AFB) and G (AFG), and also for cyclopiazonic acid (CPA), by HPLC. All *A. parasiticus* isolates were strong AFB and AFG producers, but no CPA production was detected. The *A. flavus* isolates showed to be more diversified, with 77% being atoxigenic, whereas 15% produced CPA and low levels of AFB and 8% produced the 3 groups of mycotoxins. Molecularly, two genes of the aflatoxin biosynthetic pathway, *aflD* (= *nor1*) and *aflQ* (= *ord1* = *ordA*) were tested for presence and expression (by PCR and RT-PCR, respectively). The presence of both genes did not correlate with aflatoxigenicity. *aflD* expression was not considered a good marker for differentiating aflatoxigenic from non-aflatoxigenic isolates, but *aflQ* showed a good correlation between expression and aflatoxin-production ability.

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