

Mycobiota predominant and aflatoxins content in shell and shelled Brazil nuts

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Brazil nuts (*Bertholletia excelsa* Humb. and Bonpl.) are an important product of the Brazilian Amazon. Currently, its marketing is compromised by the high incidence of aflatoxins (AF). The most known naturally occurring AF are named AFB₁, AFB₂, AFG₁, and AFG₂. This study aimed to identify the potentially aflatoxigenic mycobiota associated with shelled Brazil nuts and with the shells, and to determine which one of these fractions contributes to aflatoxins (AF) contamination, since that official method use integral Brazil nuts samples to AF test. Samples of Brazil nuts were collected from the agro forestry system production area in Amazonian rain forest, in Brazil. These samples were split in shells and shelled nuts, and the total count of *Aspergillus* spp. was analysed after sanitation (sodium hypochlorite 1% / 10 minutes) and without sanitation, by plating AFPA medium, for 7 days, at 25 °C. The isolates identified as *Aspergillus* section Flavi were plated in YES medium (5days at 25°C) for determination of the aflatoxigenic potential by agar plug technique. To analyze AF, 500 g samples were milled and were extracted with chloroform. The chromatographic analysis was performed by HPLC–FD system in an isocratic mode [Waters pump W600, Waters module autosampler W717, Fluoresce detector W2475 and column Waters X-Terra (4.6x150mm and 5µm – RP18)]. The mobile phase was water milli-Q/acetonitrile/methanol (600:150:150 v/v) and the injected volume was 5µL both to standards and samples. The average incidence of infection from *Aspergillus* spp. in sections Flavi, Nigri and Circumdati were 48%, 8% and 1%, respectively. The sanitization treatment reduced the fungi counts. There were AF production by fungi isolated from both types of samples, 30% of the samples were positive for AFB₁, AFB₂, AFG₁ and AFG₂ and 23.8% produced AFB₁, AFB₂, and AFG₁. Concerning the Brazil nuts AF analysis, it was observed that the concentration of AFB₁ and AFG₁ obtained were higher than AFB₂ and AFG₂. The AFB₁ content was 35.281 and 1.782 µg/Kg in shelled Brazil nuts and shells, respectively. AFB₂ and AFG₂ were detected only in shelled samples. The HPLC-FD presented limits of detection (LOD) and quantification (LQ) of 0.2 and 0.4 µg/kg, respectively.

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