

## 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 marketing is compromised by the high incidence of aflatoxins (AF). The most known naturally occurring AF named AFB<sub>1</sub>, AFB<sub>2</sub>, AFG<sub>1</sub>, and AFG<sub>2</sub>. This study aimed to identify the potentially aflatoxigenic mycol associated with shelled Brazil nuts and with the shells, and to determine which one of these fractions contribute aflatoxins (AF) contamination, since that official method use integral Brazil nuts samples to AF test. Sample Brazil nuts were collected from the agro forestry system production area in Amazonian rain forest, in Brazil. The samples were split in shells and shelled nuts, and the total count of *Aspergillus* spp. was analysed after sanitization (sodium hypochlorite 1% / 10 minutes) and without sanitization, by plating AFPA medium, for 7 days, at 25 °C. Isolates identified as *Aspergillus* section Flavi were plated in YES medium (5days at 25°C) for determination of aflatoxigenic potential by agar plug technique. To analyze AF, 500 g samples were milled and were extracted in chloroform. The chromatographic analysis was performed by HPLC–FD system in an isocratic mode [Waters pump W600, Waters module autosampler W717, Fluorescence detector W2475 and column Waters X-Terra (4.6x150mm 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 *F. Nigri* and *Circumdati* were 48%, 8% and 1%, respectively. The sanitization treatment reduced the fungi count. There were AF production by fungi isolated from both types of samples, 30% of the samples were positive for AFB<sub>1</sub>, AFB<sub>2</sub>, AFG<sub>1</sub> and AFG<sub>2</sub> and 23.8% produced AFB<sub>1</sub>, AFB<sub>2</sub>, and AFG<sub>1</sub>. Concerning the Brazil nuts AF analysis, it was observed that the concentration of AFB<sub>1</sub> and AFG<sub>1</sub> obtained were higher than AFB<sub>2</sub> and AFG<sub>2</sub>. The AFB<sub>1</sub> concentration was 35.281 and 1.782 µg/Kg in shelled Brazil nuts and shells, respectively. AFB<sub>2</sub> and AFG<sub>2</sub> were detected only in shelled samples. The HPLC-FD presented limits of detection (LOD) and quantification (LQ) of 0.2 and 0.4 µg, respectively.

**Keywords:** mycotoxins, food safety, fungi, *Bertholletia excelsa*