Method development for simultaneous determination of Aflatoxins B1, B2, G1,and G2 in baby food using the DLLME method and HPLC

A. Shafiee[*], H. Yazdanpanah, F. Kobarfard

Toxicology Department, School of Pharmacy, Shahid Beheshti Univercity of Tehran, Tehran, Iran

Background and Aims: Aflatoxins are toxic metabolites which contaminate a wide range of foods. Baby foods usually consist of different ingredients (such as cereal grains and dried milk) which are susceptible to aflatoxin contamination. The aim of this study was to develop an efficient HPLC method for analysis of aflatoxins in baby foods using dispersive liquid–liquid microextraction method (DLLME) for the first time.

Methods: A DLLME method was developed for extraction and clean up of aflatoxins B1, B2, G1 and G2 in baby food. The samples were extracted with 4 ml of a mixture of ACN:MeOH:H2O (60:20:20) and centrifuged at 9000 rpm for 12 min. One ml of the supernatant was transferred to a vial and 500 µl ethyl acetate added to it. The mixture was rapidly injected into a glass centrifuge tube containing 3 ml of water and centrifuged at 5000 rpm for 4 min. The supernatant ethyl acetate phase was quantitatively transferred to a vial, evaporated, reconstituted with 500 µl of MeOH/H2O (40:60) and analyzed using HPLC and fluorescent detector. The chromatographic separation was done using a C18 column eluted with isocratic mobile phase and post-column electrochemical derivatization. The method was validated using baby foods spiked with aflatoxins at 7 different levels. A spiked calibration curve was established for each aflatoxin.

Results: Validation results showed that mean recoveries of aflatoxins B1, B2, G1 and G2 in spiked baby foods ranged 71.2% to 117.8%. The variation coefficients for aflatoxins were found to be less than 20%. Limits of detection and quantification for aflatoxins ranged between 0.03-0.17 ng/g and 0.10–0.50 ng/g, respectively.

Conclusions: For the first time, a DLLME method was successfully developed for HPLC determination of aflatoxins in baby food. It was found to be accurate, precise, sensitive, efficient, easy, fast and inexpensive.

Keywords: Aflatoxins; DLLME; HPLC; Analysis