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Review Article

Aflatoxins in Food Products in Iran: a Review of the Literature

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Abstract

Context: Mycotoxins are secondary metabolites produced by certain toxigenic fungi and the most of them are aflatoxins, fumonisins, trichothecenes, ochratoxin A, patulin, and zearalenone.
Evidence Acquisition: In consideration of the consumption of certain farm products for animal feed and the prevalence of toxigenic fungi and mycotoxins in food, the present study was performed to evaluate this situation in Iran with a review of the literature using search engines. All published articles were selected using Iran Medex, Magiran, PubMed NCBI, and Google Scholar.
Results: Aflatoxins have been found in many food products in Iran.
Conclusions: It is necessary to detect aflatoxins in foods and food products as early as possible, before they enter human or animal bodies. There is a high consumption of milk and dairy products in Iran, and the proper management of animal foods can help to decrease the aflatoxins in milk.

Keywords: Aflatoxin, Foods, Iran

1. Context

Fungi are ubiquitous microorganisms in the soil and air that are often associated with spoilage and biodeterioration of foods (1-3). Mycotoxins are secondary metabolites produced by certain toxigenic fungi (4). The most important mycotoxins are aflatoxins, fumonisins, trichothecenes, ochratoxin A, patulin, and zearalenone (5-9). Aflatoxins have histopathological effects on tissues and the digestive tract based on the amount of toxin exposure (10, 11).

Aflatoxin levels in nuts, including almonds and pistachios, should not exceed 2-4 parts per billion (ppb) as mandated by importing European countries (12). Mycotoxigenic fungi naturally yield a wide range of metabolites (13). Mycotoxins such as aflatoxins play a key role in the development of food allergies, and food contaminated with aflatoxins is currently an important public health problem (14). Kazemi et al. (2008) and Ostadrahimi et al. (2014) reported high mycotoxigenic contamination in roasted salted nuts, pure samples, and bakeries (18%, 11%, and 31.5%, respectively) (15, 16). The first report on mycotoxins in Iran was published by Emami et al. in 1977, which showed different types of mycotoxin contamination of cottonseed and cottonseed cake immediately after harvest, during long and wet storage conditions (17).

Aflatoxin B1 (AFB1), Aflatoxin B2 (AFB2), Aflatoxin G1 (AFG1), and Aflatoxin G2 (AFG2), can be produced on maize, wheat, and rice (18, 19). AFM1 appears primarily in biological fluids (milk and urine) after the intake of foods contaminated with AFB1 and its subsequent transformation into AFM1 through hydroxylation (20). Ersali et al. (2009) showed that the percentage of Aflatoxin M1 (AFM1) in corn was higher than in other cow-feeds (21). Human food can be contaminated with mycotoxins, causing mycotoxicosis in humans (19, 22-25). The toxicity of mycotoxins in animals has been investigated, and the effects can be either local or disseminated (26-28). A study by Manafi (2011) showed the negative effects of toxins on biochemical parameters when used in broiler-chicken feed, especially in combined groups (29). Alinezhad et al. (2011) showed that 60.66% of trout-feed pellets infested with *A. flavus* were capable of producing AFB1 (30). *Aspergillus*, *Penicillium*, and *Fusarium* are the main mycotoxin-producing fungi in nature; however, only a few species within these genera have the capability of toxin production (such as AFB1, AFB2, AFG1, and AFG2) (18, 19, 31, 32).

Aspergillus flavus is a major fungus found in the air in warm climates (33), and other *Aspergillus* spp. (such as *parasiticus* and *versicolor*) can produce aflatoxins (34), though not all can. A study by Razzaghi-Abyaneh et al. (2006) showed that 27.5% of 66 *A. flavus* isolates were capable of producing aflatoxins (19). Corneal ulcer is a common condition caused by several microbes, including fungi, viruses, bacteria, and protozoa (35). *A. flavus* is the most common cause of fungal keratitis, and other fungi, such as *A. fumigatus*, *Fusarium* spp., and *Rhizopus* spp. can lead to fungal keratitis (36). The distribution of mycotoxins in food varies, and due to the lack of any review of this subject in Iran, the present literature review was performed using studies published through 2014.

2. Evidence Acquisition

In consideration of the use of certain farm crops for animal feed and the incidence of toxigenic fungi and mycotoxins in food, the current study was conducted to evaluate this topic in Iran, using various search engines. All articles were published in domestic and international journals and were found using Iran Medex, Magiran, PubMed NCBI, and Google Scholar. We included all results involving Iranian food products and aflatoxins. The exclusion criterion was aflatoxin contamination in imported food.

3. Results

3.1. Bread

We found only one study that measured aflatoxins in bread. This study was performed by Azadbakht et al. (2008), who examined 180 different bread samples from Lorestan province for the presence of aflatoxins. The results showed that all of the samples were contaminated with aflatoxins, and AFB1 was the most prevalent. Of the samples, 10% had a higher toxin level than allowed by national standards (28). According to some experts, the determination of aflatoxins is necessary in bread produced from maize. In one study, all 54 samples from a maize farm were contaminated with aflatoxins with a mean level of 2.79 ± 0.17 ng/g, and 14.81% of the samples had concentrations higher than 4 ng/g (37).

3.2. Cheese

As a dairy product, cheese may contain aflatoxin residues. The results of research by Kamkar et al. (2008) showed that the mean AFM1 level in cheese samples was 1.57 μg/L (38). In another study reported by Fallah et al. (2009) on 420 different types of cheese samples, 161 of the samples (76.6%) had AFM1 levels of more than 50 ng/kg (range 52.1-785.4 ng/kg) (39).

3.3. Fruit

Fruits have been assessed less often than other foods in Iran for the presence of aflatoxins. Dried vine fruit had the lowest AFB1, AFB2, AFG1, and AFG1 levels in one study (38).

3.4. Ice cream

As a milk-based product, ice cream can contain aflatoxins if they were present in the milk. In a study by Kazemi et al. (2013), 90 ice cream samples were examined for AFM1 in Guilan province, and 68.88% were found to be contaminated with AFM1 in a range of 8.4-147.7 ng/L (40).

3.5. Milk

Since milk is consumed by most people, it may produce serious problems (26, 41). Almost all studies on raw and pasteurized milk in Iran have shown aflatoxin contamination. Kamkar et al. (2011) showed that all 122 tested raw-milk samples were contaminated with AFM1 at a level of 40.01 ng/L. The AFM1 level in 14.75% of milk samples was higher than the Iranian standards (24). Sadeghi et al. (2013) showed that out of 320 raw-milk samples from pasteurized-milk factories in Kermanshah, AFM1 levels were higher than the Codex standards, with a mean level of 1.21 μg/mL (range 0.4-2.42 μg/mL), and there were significant differences between factories (42).

An investigation of 90 raw-milk samples from Guilan showed that 65.55% were contaminated with AFM1 at concentrations of 2.1-131 ng/L, using the ELISA method (40). The mean aflatoxin levels were different in studies done on milk, but aflatoxins were present in more than 80% of samples, according to Iranian national standards (43-47). Hassanzadeh Khayat et al. (1999), using HPLC/TLC, showed that 87.5% of 88 pasteurized milk samples from Mashhad had AFM1 levels between 0.105-0.525 ppb (26). In 2005-2006, 72 samples of raw and pasteurized milk were collected from Uremia, and AFM1 levels were higher than 50 ng/L in 6.25%. In the raw and pasteurized samples, respectively, the maximum AFM1 levels were 91.80 and 28.5 ng/L and the minimum levels were 4.33 and 5.1 ng/L (48).

A study by Karimi et al. (2007) showed that 5.4% of 110 pasteurized milk samples had AFM1 levels of > 0.05 μg/L (49). All pasteurized milk in Babol (northern Iran) had AFM1 contamination, with levels of > 200 ng/L in 91.5% of samples (50). AFM1 was detected in 100% of pasteurized and sterilized milk samples; 100% of the samples had toxin levels exceeding European Union regulations (50 ng/L). The mean AFM1 contamination levels of pasteurized and sterilized milk were 230.5 and 221.66 ng/L, respectively (51). The same results were observed in Kamkar et al.'s (2008) study on 52 ultra-high temperature (UHT) pasteurized milk samples, all of which showed AFM1 contamination (38).

Zanjani et al. (2015) reported that the mean AFM1 concentration in 45° raw milk samples was 11.61 ± 0.72 ng/L (range 6.3 - 23.3 ng/L) (52).

Ersali et al. (2009) examined 428 samples of raw and pasteurized milk and animal feeds in different seasons of the year, using the ELISA or TLC methods. In 38.03% of raw milk samples and 14.42% of the pasteurized milk samples, the contamination level was above the permissible limit (0.5 ppb) (21). In a study by Nemati et al. (2010), using the ELISA method, all 90 pasteurized milk samples in Ardebil were contaminated with AFM1 in a range of 2.9 - 85 ng/kg; 33% of these samples had AFM1 levels exceeding the national standards (53). Mohammadian et al. (2010) found that 4.4% of milk samples had higher AFM1 than the national standards, and they found no significant differences between raw and pasteurized milk (44). Movassagh (2011), in a study on 49 UHT milk samples from Tabriz, found that all of them had AFM1, based on ELISA (54).

A study cited by Rahimi et al. (2012) showed that 46.7% of bulk milk samples from cows, sheep, and goats were contaminated with AFM1 at 48.7 ± 23.5, 25.5 ± 15.1, and 31.6 ± 13.7 ng/L, respectively (55). The results for 132 mother's breast milk samples for the presence of AFM, using ELISA, showed that 6.06% of samples were contaminated with AFM, with a mean level of 9.45 ng/L (56). Some studies showed differences between aflatoxins in milk according to the season (46, 57), but one study found no significant differences in this regard (44).

3.6. Pistachios and Other Nuts

Certain nuts, especially pistachios, are common in Iran, and thus most researchers have focused on mycoflora and mycotoxins in these nuts. A study by Cheraghali et al. (2007) showed that out of 3,356 pistachio nut samples, AFB1 was detected in 2,852 (28.3%), with a mean level of 7.3 ± 53.2 ng/g. The AFB1 level in 1,191 samples (11.8%) was above the maximum tolerated level (MTL) for pistachio nuts in Iran (5 ng/g) (58). Shadbad et al. (2012) showed that some nuts were contaminated with aflatoxins in Tabriz, based on the HPLC method. The incidence rates of aflatoxin were 14.29% (almonds), 76.92% (walnuts), 25% (apricots), 33.33% (peanuts), 7.69% (hazelnut), 53.13% (pistachios), and 14.29% (cashews); sunflower and sesame seeds showed no aflatoxins (59). Amiri et al. (2013) showed that 100% of 80 nut samples (peanuts, almonds, walnuts, and hazelnuts) were contaminated with aflatoxin in ranges of 0.016 - 15.74 μg/kg. The highest aflatoxin level was found in peanuts (1.61 μg/kg) and the lowest was in almonds (0.27 μg/kg) (60). Another study using the ELISA technique on 167 samples of figs, almonds, hazelnuts, walnuts, pistachios, and sunflower seeds showed that 59.9% were contaminated with aflatoxins at a mean level 1.12 μg/kg. Aflatoxin levels in all samples were lower than the Iranian national standards (61).

In a study of peanuts by Khorasgani et al. (2013), the mean aflatoxin level of 1.12 μg/kg was lower than the Iranian national standards (62). In a study by Khorasgani et al. (2013) on peanuts in Shakeri et al. (2014) found AFB1, AFB2, AFG1, AFG2, and total aflatoxin levels of 8.32, 5.64, 3.07, 1.71, and 10.38 ng/kg, respectively, in 10% of 80 nut samples (pistachios, almonds, hazelnuts, and walnuts). Pistachio samples had the highest aflatoxin contamination levels (63). A study by Rezaei et al. (2014) on aflatoxin contamination using ELISA in figs, almonds, hazelnuts, walnuts, pistachios, and sunflower seeds showed that 96.5% of 200 samples were contaminated with aflatoxins, with a mean level of 1.68 μg/kg (range 0 - 6 μg/kg) (64). Ostadrahimi et al.'s (2014) study of raw and salted nuts showed that 90% of salted/roasted walnut samples and 58.6% of pistachio samples were contaminated with aflatoxins. The mean aflatoxin concentration in all samples was 19.88 ± 19.41 μg/kg (15).

3.7. Rice

Rice is cultured in provinces of northern Iran. In a study performed by Zaboli et al. (2011) to determine the correlation between AFB1 and *Aspergillus* in rice, 30 samples showed no differences in AFB1 levels in new and old rice (65). Using the HPLC method, Amanloo et al. (2014) showed that 27.6% of 123 imported rice samples were contaminated with aflatoxins; AFB1 was the most prevalent, followed by AFG1, AFB2, and AFG2 (66). Karajibani et al. (2013) reported that AFB1 and AFB2 in 33% and 47% of yellow and white rice samples were contaminated with 0.34 and 0.58 ppb, respectively. It was also detected 13% and 21% following samples also had AFB2 contamination at 0.06 and 0.08 ppb, respectively (67). Khorasgani et al. (2015) showed that the highest concentrations of AFB1 and total AFs in imported rice samples were 2.350 and 2.704 ng/g, respectively (68).

3.8. Sesame Seeds

Sesame seed belongs to the *Pedaliaceae* family, and was studied for mycotoxins by Hosseinnia et al. (2014) in Khorasan Razavi province. Aflatoxin was found in 50% of the samples, but at low levels; only 1.9% of samples had levels exceeding the standards (69).

3.9. Yogurt

Yogurt is a dairy product consumed in various forms throughout Iran. One study showed that 100% of 50 yogurt samples were contaminated with AFM1 (70). Fallah et al. (2010) reported that only 66.1% of 68 yogurt samples contained AFM1 (71). Other studies showed that the contamination rate of AFM1 in yogurt was 80% - 98.33% (72). AFM1 was found in all commercial and traditional yogurt samples in Guilan Province in northern Iran, at concentration levels in the range of 4.2 - 78.9 ng/kg (73). Behnamipour et al. (2014) showed that all 28 yogurt samples in Qom were contaminated with AFM1 in the range of 5–36 ng/kg, with a mean of 13.55 ng/kg (43).

3.10. Wheat

Wheat flour is used in food products such as breads, cakes, and biscuits, and can be infested with fungi at farms, which can cause contamination of these foods with mycotoxins. Aflatoxins were identified in 2.54% of wheat samples (mean value of 3.12 ppb). AFB1 and AFG1 were found in 2.54% and 3.39% of samples, respectively. Contamination of wheat samples with aflatoxins was not related to *A. flavus*. The aflatoxin rate did not exceed 30 ppb in any wheat samples in one study (74). Taheri et al. (2012) analyzed 200 wheat-flour samples for aflatoxin contamination using the HPLC method, and the detection rates in summer and winter, respectively, were 70% and 99%, at 0.82 and 1.99 ng/g (75). Almost all samples of Kashkineh, a wheat product, were free of any AFB1, AFB2, AFG1, or AFG2 in one study (76) (Table 1).

4. Conclusions

Research carried out in different regions and provinces of Iran has shown many genera and species of toxigenic fungi, and aflatoxins have been found in different types of food, air, and equipment. Animal feed is an important route for the entrance of mycotoxins into human tissues, and thus certain management activities are important in order to decrease mycotoxin levels in foodstuffs. Based on the levels of milk and dairy consumption in Iran, many control processes are aimed at decreasing mycotoxin production in milk and using safe animal feeds. Farm-management and food-storage practices are effective at decreasing food-processing times, and these efforts can prevent or minimize toxin formation in agriculture, industry, and food-product manufacturing in order to improve human and animal health. The routine detection of mycotoxins in food and food products for human and animal consumption should be performed as early as possible, before these toxins enter human or animal bodies.

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Footnotes

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Table 1. Types of Aflatoxins Isolated in Iran

Fungal Source	Source	Toxins					% of Infection	Season	Reference
		AFM1	AFB1	AFB2	AFG1	AFG2			
-	Pasteurized milk	0105 - 0525 ppb[a]	-	-	-	-	-	-	(26)
-	Wheat	-	1.53 ppb[a]	2.75 ppb[a]	-	-	2.54	-	(74)
A. flavus	Farm soil	-	0.08-2.29 μg/mL[d]	-	-	-	-	-	(19)
A. flavus	Farm soil	-	0.00-1.69 μg/mL[c]	-	-	-	-	-	(19)
A. parasiticus	Farm soil	-	1.85-2.77 μg/mL[d]	-	-	-	-	-	(19)
A. parasiticus	Farm soil	-	0.41-1.05 μg/mL[c]	-	-	-	-	-	(19)
A. nomius	Farm soil	-	0.15-1.18 μg/mL[d]	-	-	-	-	-	(19)
A. nomius	Farm soil	-	0.00-0.51 μg/mL[c]	-	-	-	-	-	(19)
-	Pasteurized milk	114.35 ng/L	-	-	-	-	-	Winter	(50)
-	Pasteurized milk	230.2 ng/L	-	0.4 ng/g	-	-	-	Summer	(50)
-	Pistachios	–	2 ng/g[b]	2.44 μg/kg[a]	2 ng/g	0.4 ng/g	-	-	(58)
-	Bread	-	22.53 μg/kg[a]	-	0.19 μg/kg[a]	-	100	-	(28)
-	Pasteurized milk	12.65 ± 17.76 ng/L[b]	-	-	-	-	96.88	All	(44)
-	Raw milk	12.43 ± 17.53 ng/L[b]	-	-	-	-	94.17	All	(44)
-	UHT milk	111.45 ± 10.42 ng/L[b]	-	-	-	-	100	Summer/autumn	(54)
-	Pasteurized milk	43.2 ± 22.8 ng/L[b]	-	-	-	-	60	Winter	(55)
-	Pasteurized milk	21.3 ± 9.6 ng/L[b]	-	-	-	-	40	Spring	(55)
-	Pasteurized milk	30.2 ± 9.2 ng/L[b]	-	-	-	-	33.3	Summer	(55)
-	Pasteurized milk	41.3 ± 27.2 ng/L[b]	-	-	-	-	26.7	Autumn	(55)
-	White cheese	148.5 ± 52. ng/L[b]	-	-	-	-	66.7	Winter	(55)
-	White cheese	78.9 ± 48.6 ng/L[b]	-	-	-	-	75	Spring	(55)
-	White cheese	70.8 ± 22.2 ng/L[b]	–	-	-	-	66.7	Summer	(55)
-	White cheese	138.4 ± 42.4 ng/L[b]	-	-	-	-	33.3	Autumn	(55)
-	Pasteurized milk	60.37 ± 34.66 ng/L[b]	-	-	-	-	-	Spring	(57)
-	Pasteurized milk	61.68 ± 27.57 ng/L[b]	-	-	-	-	-	Summer	(57)
-	Peanuts	-	0.37 - 3.78 μg/kg[a]	-	-	-	45.12	-	(59)
-	Peanuts	-	-	0.2-42.05 μg/kg[a]	-	-	14.32	-	(59)
-	Peanuts	-	-	-	0.11 - 225.8 μg/kg[a]	-	18.25	-	(59)
-	Peanuts	-	-	-	-	0.04 - 15.2 μg/kg[a]	22.31	-	(40)
-	Walnuts	-	15.4 - 35.1 μg/mL[b]	4 - 8.1 μg/mL[c]	1.4 - 8.2 μg/mL[c]	-	76.92	-	(62)
-	Pistachios	-	9.5 - 43.8 μg/mL[b]	0.9-9.4 μg/mL[c]	19.7 μg/mL[c]	5.1 μg/mL[c]	53.13	-	(62)
-	Cashews		18.3 μg/mL[b]	2.7 μg/mL[c]	-	7.1 μg/mL[c]	14.29	-	(62)
-	Raw milk	2.1 - 131 ng/L[c]	-	-	-	-	65.55	Autumn/winter	(62)
-	Ice cream	8.4 - 147.7 ng/L[b]	-	-	-	-	68.88	-	(72)
-	Pistachios	-	7.34 ± 6.77 ppb[a]	4.19 ppb[a]	3.99 ± 3.94 ppb[a]	0.83 ppb[a]	10	-	(37)
-	Maize	-	2.79 ± 0.17 ng/g[a*]	-	-	-	100	Spring	(63)
-	Almonds	-	3.43 ppb[a]	-	2.36 ppb[a]	-	2.5	-	(63)
-	Hazelnuts	-	-	-	0.17 ppb[a]	-	1.25	-	(63)
-	Walnuts	-	12.23 ± 10.06 ppb[a]	7.08 ppb[a]	3.88 ppb[a]	1.71 ± 1.81 ppb[a]	6.25	-	(63)
-	Feed materials	-	1.83 μg/kg[b*]	-	-	-	45	Autumn	(64)
-	Rice	-	1.02 ng/g-	-	-	0.1 μg/kg[a]	27.6	-	(66)
-	Raw walnuts	-	6.51 ± 9.4 μg/kg[b*]	-	-	-	2.3	-	(15)
-	Raw pistachios		14.4 ± 8.4 μg/kg[b*]	-	-	-	17.3	-	(15)
-	Raw peanuts	-	3.03 ± 8.6 μg/kg[b*]	-	-	-	90.69	-	(15)
-	Salted pistachios	-	22.2 ± 20.2 μg/kg[b*]	-	-	-	55.17	-	(15)
-	Salted peanuts	-	17.99 ± 18.7 μg/kg[b*]	-	-	-	15.15	-	(15)
-	White rice	-	0.16 - 1.82 (0.62 ± 0.43) ppb[a]	0.00 - 0.70 (0.09 ± 0.18) ppb[a]	-	-	47	Spring	(67)

-	White rice	-	0.08 - 2.36 (0.87 ± 0.66) ppb[a]	0.00 - 0.26 (0.07 ± 0.08) ppb[a]	-	-	47	Summer	(67)
-	White rice	-	0.40 - 0.43 (0.40 ± 0.42) ppb[a]	0.00 - 0.09 (0.01 ± 0.03) ppb[a]	-	-	47	Autumn	(67)
-	White rice	-	0.07-1.08 (0.45 ± 0.28) ppb[a]	0.00 - 0.96 (0.01 ± 0.02) ppb[a]	-	-	47	Winter	(67)
-	Yellow rice	-	0.11-0.87 (0.430 ± 0.26) ppb[a]	0.00 - 0.07 (0.03 ± 0.03) ppb[a]	-	-	21	Spring	(67)
-	Yellow rice	-	0.07 - 0.88 (0.46 ± 0.27) ppb[a]	0.00 - 0.11 (0.05 ± 0.04) ppb[a]	-	-	21	Summer	(67)
-	Yellow rice	-	0.10 - 0.79 (0.34 ± 0.22) ppb[a]	0.00 - 0.17 (0.01 ± 0.04) ppb[a]	-	-	21	Autumn	(67)
-	Yellow rice	-	0.01 - 0.13 (0.07 ± 0.05) ppb[a]	0.00 - 0.01 (0.002 ± 0.004) ppb[a]	-	-	21	Winter	(67)