



Fungal Contamination of Feed Material Manufactured in Iran with Emphasis on Its Importance in Safety of Animal Origin Foods

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Abstract

Background: Mycotoxins are a group of structurally diverse substances produced by a wide range of moulds. These fungal secondary metabolites, whose presence in feed is unavoidable, can impose hazards to human and animal health. Continuous concern about potential effects of mycotoxins in the human diet has led to an increasing interest in researches about fungi and mycotoxin production. The aim of this work was to assess fungal contamination of feed material manufactured in Iran with emphasis on its importance in safety of animal origin foods.

Methods: Samples included 16 poultry and 15 aquatic feed which collected from 4 and 3 factories, respectively in Markazi province, Iran, in 2012. Then, standard mould count procedure was carried out to determine fungal contamination levels in the samples. SPSS (version 16) software was applied to statistical analysis.

Results: According to the results, 6.7% of aquatic and 31.3% of poultry feed samples had fungal contamination. The mean fungal contamination level of poultry feed and aquatic feed samples were $6.4 \times 10^4 \pm 1.12 \times 10^5$ CFU/g and $1.18 \times 10^3 \pm 2.77 \times 10^3$ CFU/g, respectively.

Conclusion: This study showed a need for control of fungal contamination in aquatic and poultry feed in Iran. The condition of poultry feed is more problematic than that of aquatic feed, indicating a need to more local work to have least fungal contamination.

Introduction

When animal feed materials are contaminated by moulds, there is a significant risk of their contamination with the secondary metabolites of these fungi called "Mycotoxins". Thus, fungal contamination should be examined for feeds of local areas in different countries and hence the results could be practical for the studied regions (Čonková et al., 2006; Kabak et al., 2006). Diverse mycotoxins produced by several fungi species are mainly aflatoxin, ochratoxin A, fumonisins, deoxynivalenol, zearalenone, etc. which among them aflatoxins are of much significance. The most important aflatoxin producers fungi occurring naturally in feed

and food materials are *Aspergillus flavus*, *A. parasiticus* and, to a lesser extent, *A. nomius* (Murphy et al., 2006; Neal et al., 1998; Pitt, 2000). Aflatoxin M1 is the metabolite of AFB1 and exists mainly in milk of farm animals ingesting feed contaminated with AFB1 (Montaseri et al., 2014). Since mycotoxin producer fungi are widely distributed in soil, air and plant materials (Astoreca et al., 2011), there is a serious hygienic risk about fungal contamination of industrial animal feed endangering safety of food of animal origin especially milk and meat.

Controlling fungal contamination should be considered in animal nutrition to decrease mycotoxin concentration in human diet. By regard to the importance of this issue,

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the aim of this work was to assess fungal contamination of feed material manufactured in Iran with emphasis on its importance in safety of animal origin foods.

Materials and methods

Samples collection

Samples included 16 poultry feed and 15 aquatic concentrated feed samples which collected from 4 and 3 factories, respectively in Markazi province, Iran, in 2012. Markazi province, is one of the most important industrial regions of Iran in which several industrial factories are located.

Fungal culture

Total fungal contamination analysis was performed according to ISO (2008). In detail, 20 g of each sample were homogenized for 5 min in 180 ml peptone-water. Decimal dilutions in 9% (w/v) NaCl solution were prepared and inoculated in dichloran 18% mass fraction glycerol agar medium containing yeast extract (5 g), glucose (10 g), chloramphenicol (0.1 g), agar (12-15 g), $\text{PO}_4\text{H}_2\text{K}$ (1 g), $\text{MgSO}_4\cdot\text{H}_2\text{O}$ (0.5 g), dichloran (2, 6-dichloro-4-nitroaniline)

(0.002 g), glycerol (220 ml), digestion enzyme of casein (5 g) and distilled water (up to 1 l).

Plates were incubated for 5 days at 30 °C, after which colonies were counted and then the data were recorded.

Statistical analysis

SPSS (version 16) software was applied to statistical analysis.

Results

According to the results, 6.7% of aquatic and 31.3% of poultry feed samples had fungal contamination. The mean fungal contamination level of poultry feed and aquatic feed samples were $6.4\times 10^4\pm 1.12\times 10^5$ CFU/g and $1.18\times 10^3\pm 2.77\times 10^3$ CFU/g, respectively.

The contamination levels in 1 of 3 aquatic feed factories and 2 of 4 poultry feed factories were above acceptable level ($<10^4$ CFU/g).

As shown in Table 1 and Table 2, significance differences was observed between fungal contamination level of different feed types ($p<0.05$).

Table 1: Fungal contamination of aquatic feed produced in different factories in Markazi province of Iran

Factory number	Fungal contamination (Mean±SD)	Samples (%) with $>10^4$ (CFU/g)*
1	$7.86\times 10^2\pm 6.32\times 10$	0
2	$3.34\times 10^2\pm 2.07\times 10^2$	0
3	$3.12\times 10^3\pm 4.43\times 10^3$	20
Mean	$1.18\times 10^3\pm 2.77\times 10^3$	6.7

* Acceptable level is $<10^4$ (CFU/g)

Table 2: Fungal contamination of poultry feed produced in different factories in Markazi province of Iran

Factory number	Fungal contamination (Mean±SD)	Samples (%) with $>10^4$ (CFU/g)*
1	$1.07\times 10^2\pm 62.5\times 10$	0
2	$2.62\times 10^3\pm 4.92\times 10^3$	0
3	$4.71\times 10^4\pm 8.86\times 10^4$	25
4	$2.0625\times 10^5\pm 1.29\times 10^5$	100
Mean	$6.4019\times 10^4\pm 1.12\times 10^5$	31.3

* Acceptable level is $<10^4$ (CFU/g)

Discussion

Mycotoxins have adverse effects on humans, animals and crops, resulting in illness and economic losses (Rosa et al., 2006). This study showed that 6.7% of aquatic and 31.3% of poultry feed samples had fungal contamination. Moulds are known as the most frequent contaminant of animal feed. According to International Organization for Standardization (2008), total fungal count of food and animal feed

should be below 10^4 CFU/g. Based on Table 1 and Table 2, among 7 feed factories, 3 of them had the samples with fungal contamination counts above standard level. Such situation seems to be warning from hygienic aspects. It is possible that the factories with high level of contamination have not sufficient sanitation facilities or its employers may have not enough training. Feed contamination comes from different sources, such as failure to comply with health codes or contamination of the production line. Feeds

may be contaminated by fungi during milling process. Therefore, in addition to the feed, the processing pathway will also increase the fungal pollution. Feeds contamination depends on different factors such as human error, production practices and handling procedures in the feed mill, during transport and on the farm. In feed mills, fungal contamination may be retained at various points along the production line, contaminating successive batches of meal as they are processed (Kabak et al., 2006; Murphy et al., 2006; Pitt, 2000).

Recently, Mahmoudi (2014) reported the considerable concentrations of Zearalenone in food of animal origin produced in North-West of Iran. Among 210 samples of milk, liver and meat, 92 of them (43.8%) were contaminated with this mycotoxin. The significantly higher contamination rate was observed in autumn compared to summer. Also, several researches carried out in Iran and the other countries have indicated high incidence rate of aflatoxin M₁ in milk of farming animals (Bakirci, 2001; Fallah, 2010a; Fallah, 2010b; Fallah et al., 2011; Ghazani, 2009; Prandini et al., 2009), resulting from considerable presence of mycotoxin producer fungi in consumed feed of such animals.

In a study in Canada, mycotoxins were found in 18% of feed samples during 1982-1994 (Abramson et al., 1997). Similar finding has been reported by Jaimez et al. (2004) who worked on the occurrence of fungal contamination and mycotoxin residue in animal feed produced in Spain. Previous investigation about fungal contamination of animal feed in the other province of Iran were in accordance with our finding (Fani et al., 2013; Rezaei et al., 2013a; Rezaei et al., 2013b; , Rezaei et al., 2014a; Rezaei et al., 2014b).

Conclusion

Considering the results obtained from the survey and the previous researches carried out in Iran, it is concluded that there is a clear need to control fungal contamination of animal feeds in Iran by paying more attention to practical decisions to prevent transfer of these toxins to animals or to animal products.

This study measured the fungal contamination in poultry and aquatic feed in Markazi province of Iran. As poultry and aquatic products are the main protein sources for humans, their safety should be considered as a serious issue. This study showed a need for control of fungal contamination in aquatic and poultry feed in Iran. The condition of poultry feed is more problematic than that of aquatic one, indicating a need to more local work to have least fungal contamination.

Conflicts of interest

There is no conflict of interest.

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References

- Abramson D., Mills J., Marquardt R., Frohlich A. (1997). Mycotoxins in fungal contaminated samples of animal feed from western Canada, 1982-1994. *Canadian Journal of Veterinary Research*. 61: 49.
- Astoreca A.L., Dalcero A.M., Pinto V.F., Vaamonde G. (2011). A survey on distribution and toxigenicity of *Aspergillus* section Flavi in poultry feeds. *International Journal of Food Microbiology*. 146: 38-43.
- Bakirci I. (2001). A study on the occurrence of aflatoxin M₁ in milk and milk products produced in Van province of Turkey. *Food Control*. 12: 47-51.
- Čonková E., Laciaková A., Štyriak I., Czerwiecki L., Wilczyńska G. (2006). Fungal contamination and the levels of mycotoxins (DON and OTA) in cereal samples from Poland and East Slovakia. *Czech Journal of Food Sciences*. 24: 33-40.
- Fallah A.A. (2010a). Aflatoxin M₁ contamination in dairy products marketed in Iran during winter and summer. *Food Control*. 21: 1478-1481.
- Fallah A.A. (2010b). Assessment of aflatoxin M₁ contamination in pasteurized and UHT milk marketed in central part of Iran. *Food and Chemical Toxicology*. 48: 988-991.
- Fallah A.A., Rahnama M., Jafari T., Saei-Dehkordi S.S. (2011). Seasonal variation of aflatoxin M₁ contamination in industrial and traditional Iranian dairy products. *Food Control*. 22: 1653-1656.
- Fani A., Rezaei M., Moini A., Fani P., Mirzajani P., Malekiran A.A., Rafeie M., Atabak N. (2013). Aflatoxin occurrence in nuts consumed in Arak, Iran. *International Journal of Food Nutrition and Safety*. 4: 91-97.
- Ghazani M.H. (2009). Aflatoxin M₁ contamination in pasteurized milk in Tabriz (northwest of Iran). *Food and Chemical Toxicology*. 47: 1624-1625.
- International Organization for Standardization (ISO). (2008). Microbiology of food and animal feeding stuffs-Horizontal method for the enumeration of yeasts and moulds-Part 2: Colony count technique in products with water activity less than or equal to 0.95. No. 21527-2.
- Jaimez J., Fente C.A., Franco C.M., Cepeda A., Vázquez B.I. (2004). A survey of the fungal contamination and presence of ochratoxin A and zearalenone on Spanish feed and raw materials. *Journal of the Science of Food and Agriculture*. 84: 832-840.
- Kabak B., Dabson A.D., Var I. (2006). Strategies to prevent mycotoxin contamination of food and animal feed: A review. *Critical Reviews in Food Science and Nutrition*. 46: 593-619.
- Mahmoudi R. (2014). Occurrence of zearalenone in raw animal origin food produced in North-West of Iran. *Journal of Food Quality and Hazards Control*. 1: 25-28.
- Montaseri H., Arjmandtalab S., Dehghanzadeh G., Karami S., Razmjoo M.M., Sayadi M., Oryan A. (2014). Effect of production and storage of probiotic yogurt on aflatoxin M₁ residue. *Journal of Food Quality and Hazards Control*. 1: 7-14.
- Murphy P.A., Hendrich S., Landgren C., Bryant C.M. (2006). Food mycotoxins: an update. *Journal of Food Science*. 71: 51-65.
- Neal G.E., Eaton D.L., Judah D.J., Verma A. (1998). Metabolism and toxicity of aflatoxins M₁ and B₁ in human-derived in vitro system. *Toxicology and Applied Pharmacology*. 151: 152-158.
- Pitt J.I. (2000). Toxigenic fungi: which are important. *Medical Mycology*. 38: 17-22.
- Prandini A., Tansini G., Sigolo S., Filippi L., Laporta M., Piva G. (2009). On the occurrence of aflatoxin M₁ in milk and dairy products. *Food and Chemical Toxicology*. 47: 984-991.
- Rezaei M., Akbari Dastjerdi H., Jafari H., Farahi A., Shahabi A., Javdani H., Teimoory H., Yahyaei M., Malekiran A.A. (2014a). Assessment of dairy products consumed on the Arak market as determined by heavy metal residues. *Health*. 6: 323-327.

- Rezaei M., Karimi F., Yahyaei M., Javdani H., Shahabi A., Farahi A. (2013a). A survey of microbial total count and prevalence of *Escherichia coli* in raw milk in Markazi Province, Iran. *Research Opinions in Animal and Veterinary Sciences*. 12: 474-747.
- Rezaei M., Mohammadpourfard I., Malekiran A. A., Ghasemikhah R., Sayadi M. (2014b). Aflatoxin contamination of feed materials in Qom province, Iran. *Iranian Journal of Toxicology*. 8: 1025-1027.
- Rezaei M., Shariatifar N., Jahed Khaniki G., Javadzadeh M. (2013b). Nitrite in hamburgers in Arak, Iran. *Food Additives and Contaminants: Part B*. 6: 285-288.
- Rosa C.A., Ribeiro J.M., Fraga M.J., Gatti M., Cavaglieri L.R., Magnoli C.E., Dalcero A.M., Lopes C.W. (2006). Mycoflora of poultry feeds and ochratoxin-producing ability of isolated *Aspergillus* and *Penicillium* species. *Veterinary Microbiology*. 113: 89-96.