Aflatoxin Situation Analysis – Summary

Background

According to the Food and Agriculture Organization (FAO), around 25% of the world's food crops are contaminated with mycotoxins at levels that are relevant to food safety. However, more recent studies suggest that between 60% and 80% of crops may contain detectable levels of mycotoxins. Mycotoxins are secondary metabolites produced by filamentous fungi (Molds) that enter the food chain through the contamination of agricultural commodities intended for animal feed and food production. Aflatoxins are widely recognized as the most hazardous group of mycotoxins. They are produced mainly by toxigenic strains of Aspergillus flavus and Aspergillus parasiticus, as well as by less common species such as A. *nomius*, A. *pseudonomius*, A. *bombycis*, A. *ochraceoroseus*, A. *parvisclerotigenus*, A. *pseudotamarii*, A. *rambellii,* and A. *toxicarius.*

Based on their chemical structure, aflatoxins belong to the difuranocoumarin group. More than 20 aflatoxins have been identified; however, the most relevant are aflatoxin B1 (AFB1), aflatoxin B2 (AFB2), aflatoxin G1 (AFG1), aflatoxin G2 (AFG2), and aflatoxin M1 (AFM1) (IARC, 2012; Zain, 2011). The letters "B" and "G" refer to blue and green fluorescence under UV light, while "M" denotes milk.

AFM1 is a 4-hydroxylated metabolite of AFB1 and can be detected in the milk of dairy cattle fed AFB1-contaminated feed. Approximately 0.3–6.2% of ingested AFB1 is converted into AFM1, depending on factors such as breed, milk yield, stage of lactation, and environmental conditions. If contaminated feed is withdrawn, AFM1 levels gradually decline and are usually undetectable within 2–3 days. Following prolonged exposure to high levels of AFB1, this process takes 5–7 days.

Among all aflatoxins, AFB1 is considered the most potent carcinogenic and genotoxic compound and is also the form most frequently detected in food and feed. It is strongly associated with hepatocellular carcinoma (HCC); consequently, the International Agency for Research on Cancer (IARC) has classified AFB1 as a Group 1 human carcinogen. Although AFM1 is less toxic than AFB1, it is still classified as a human carcinogen by IARC. AFM1 contamination in milk is one of the most significant aflatoxin-related food safety concerns, because milk and dairy products are essential components of the human diet, especially for infants and children, who are particularly vulnerable due to their higher intake-to-body weight ratio and lower detoxification capacity. Elderly individuals are also considered more sensitive due to weakened immune and metabolic functions. Furthermore, AFM1 is relatively heat stable. Common dairy processing technologies, including pasteurisation (72–75 °C), ultra-high temperature treatment (120-150 °C), and membrane separation techniques, do not significantly reduce or eliminate AFM1 contamination in milk and dairy products. Consequently, systematic surveillance and control of mycotoxins in food and feed are necessary to minimise health risks and maintain food safety standards.

Overview of Aflatoxin Regulation in Hungary

Aflatoxins are strictly regulated worldwide due to their significant public health relevance. In Hungary, the regulatory framework fully aligns with the harmonised legal standards of the European Union (EU). The regulatory framework establishes distinct maximum permitted levels for aflatoxins in feed and food.

Directive 2002/32/EC. regulates the undesirable substances, also the aflatoxins in animal feed. The maximum permitted concentration of aflatoxin B1 is 0.02 mg/kg for most complete feeds, while a stricter limit of 0.005 mg/kg applies to feed for lactating animals, due to the risk that aflatoxin B1 is metabolised into aflatoxin M1, which can be excreted in milk.

The food safety legislation in the European Union has long been based on Regulation (EC) No 1881/2006, which set maximum levels for various contaminants in food, including mycotoxins. The Food Contaminants Regulation (EU) 2023/915 has replaced Regulation (EC) No 1881/2006, after 17 years which had previously established maximum levels for contaminants in food. Subsequently, Regulation (EU) 2024/1756 further refined and consolidated these provisions, contributing to a more coherent and modern regulatory framework. Regarding aflatoxins, the updated regulatory system did not introduce fundamentally new maximum limits. Instead, it clarified product classifications, particularly the distinction between raw materials and finished products, and refined the conditions under which specific limits apply. Notably, the differentiation between products intended for further processing and those ready for direct consumption is now more explicit, which is of considerable practical importance for regulators and industry stakeholders, respectively. In certain cases, maximum levels also apply to processed foods, depending on their composition and the proportion of the relevant ingredient. The maximum levels of aflatoxins also apply to processed foods if they consist of at least 80% of the corresponding product. For example, important clarification concerns aflatoxin M1, with the new regulation extending the scope of the maximum limit of 50 ng/kg to cover not only raw milk but also milk intended for direct human consumption (drinking milk). This clarification ensures a more consistent application of safety standards across different categories of milk products. The table below shows the maximum permitted levels of aflatoxins (B1, M1) in feed and milk, as set out in Regulation (EU) 2023/915.

Food categories	Maximum level of aflatoxins (µg/kg)	
	B1	M 1
Raw milk, heat-treated milk and milk for the manufacture of milk-based products	-	0,050
Infant formulae, follow-on formulae and young-child formulae	-	0,025
Baby food and processed cereal-based food for infants and young children	0,10	-
Food for special medical purposes intended for infants and young children	0,10	0,025

Table 1: Maximum permitted levels of aflatoxins in selected food categories.

This is one of the strictest regulatory frameworks for aflatoxin M1 (AFM1) in milk worldwide. In the Visegrád Group (V4, comprising Hungary, the Czech Republic, Slovakia, and Poland), there are no national differences, as the limits are harmonised under EU legislation. These AFM1 thresholds in milk are significantly stricter than those applied in countries such as Serbia (0,25 µg/kg), the United States, and China (0,5 µg/kg).

Authorities Responsible for Legislation, Investigation, and Supervision

The Hungarian regulatory framework for controlling aflatoxins is the result of collaborative efforts by several institutional entities. Those responsible for legislation and enforcement include the Ministry of Agriculture, the National Food Chain Safety Office (NÉBIH) and the National Tax and Customs Administration (NAV), and National Centre for Public Health and Pharmacy (NNGYK).

The Ministry of Agriculture is responsible for enacting legislation and providing strategic policy direction. It develops national food safety regulations in line with European Union requirements and coordinates the activities of the relevant authorities. The Ministry ensures that

EU requirements, such as mycotoxin limits, are properly incorporated into the national legal framework, ensuring that Hungarian regulations are fully aligned with those of the European Commission. The Ministry also identifies areas requiring increased monitoring and provides strategic policy direction. For example, maize is considered a high-risk commodity in drought years due to the increased likelihood of aflatoxin contamination. Additionally, the Ministry establishes national monitoring programmes by defining the scope and structure of sampling and testing activities. This includes targeted products, such as milk, feed and imported goods, as well as the frequency and intensity of controls. Furthermore, the Ministry plays a key role in crisis management. In the event of serious contamination, such as the detection of aflatoxin M1 in milk, it oversees the implementation of sector-specific measures, ensures coordinated action among relevant authorities and stakeholders, and supports effective risk communication.

The National Food Chain Safety Office (NÉBIH) is responsible for the control, testing, and enforcement of regulations relating to aflatoxins. Its activities include sampling maize, animal feed, milk, dairy products, and imported foods. The office conducts laboratory analyses to detect aflatoxins B1 and M1. If regulatory limits are exceeded, it takes action, such as ordering the seizure, recall, and withdrawal of products from the market. NÉBIH also carries out food chain supervision inspections and operates risk-based monitoring programmes. Its overall mission is to ensure food safety through systematic sampling, testing, and enforcement measures. Within the European Union, Member States use the Rapid Alert System for Food and Feed (RASFF) to notify each other when unsafe food or feed products are identified or when contamination levels exceed legal limits. The system was established under Article 50 of Regulation (EC) No 178/2002. As the national contact point, NÉBIH evaluates and forwards notifications concerning incidents relevant to Hungary or detected within the country, including alerts related to aflatoxins. In addition, Commission Regulation (EC) No 669/2009 introduces enhanced controls on products imported from third countries, with a particular focus on aflatoxin contamination. These controls are carried out regularly at the EU's external borders.

The National Tax and Customs Administration of Hungary (NAV) contributes to aflatoxin control through border and customs inspections. Its role is to inspect food and feed products imported from outside the European Union, with particular attention to high-risk commodities such as nuts and cereals. In cases of suspected risk, NAV is authorised to detain shipments and initiate sampling procedures. During inspections, it cooperates with the competent food chain safety authorities, which perform laboratory analyses and assess compliance with regulatory

limits. If irregularities are identified, contaminated shipments may be rejected or destroyed, and legal proceedings may be initiated against the importer if necessary.
The National Centre for Public Health and Pharmacy (NNGYK) is responsible for the scientific evaluation of public health risks associated with aflatoxins in Hungary. The institution plays a key role in toxicological assessment and in determining the potential health impacts of aflatoxin exposure through food consumption.
In addition, the NNGYK contributes to the health-based establishment and justification of regulatory limit values, ensuring that food safety standards are supported by scientific evidence. The centre also provides expert support for national decision-makers and participates in public health and epidemiological evaluations related to food safety hazards.
Overall, the NNGYK serves as the main scientific authority in Hungary for assessing the health risks of aflatoxins and supporting evidence-based risk management and regulatory decisions.

Overall, the system operates in a coordinated way: the ministry regulates, NÉBIH inspects and enforces, NAV filters imports at the borders, and NNGYK provides scientific risk assessment, together ensuring food safety. It is important to emphasize that the primary responsibility for ensuring the safety of food and feed lies with food business operators under European Union law. Food safety must be ensured throughout the entire food chain — from farm to table — in compliance with applicable legislation and maximum limits, through the implementation of Good Agricultural Practices (GAP) and Good Hygiene Practices (GHP), as laid down in Regulation (EC) No 178/2002.

National statistics, trends

Aflatoxin contamination in Hungary is strongly influenced by climatic conditions, with increased levels typically observed during hot and dry years. Maize production represents the most important risk factor, as aflatoxin AFB1 contamination in feed can enter the food chain and subsequently appear in animal-derived products, particularly milk in the form of aflatoxin M1 (AFM1). According to nationwide monitoring programmes conducted by the National Food Chain Safety Office (NÉBIH), thousands of samples analysed annually, the majority of which comply with the relevant regulatory limits. Non-compliance is primarily associated with feed and raw agricultural commodities, while strict control measures ensure that contamination in dairy products rarely reaches levels of public health concern. The most severe contamination event occurred in 2012–2013, when extreme drought conditions led to elevated AFB1 levels in animal feed across Hungary. This resulted in a temporary increase in AFM1 concentrations in milk. According to NÉBIH reports, approximately 7% of raw milk samples exceeded the legal limit during 2013; however, contaminated batches were identified and removed from the food chain before reaching consumers. The main source of contamination was drought-affected maize used in dairy cattle feed, with the highest risk observed in southern regions of the country. Following this event, the Hungarian monitoring and prevention system was significantly strengthened through more intensive sampling, stricter feed control, enhanced traceability, and targeted monitoring of high-risk and imported feed materials. As a result, exceedances of regulatory limits have become rare and are mainly associated with exceptionally warm and dry years. More detailed integrated monitoring and research databases, comprising approximately 2,600 analytical results, confirm that AFM1 contamination in milk shows a strong climate-dependent pattern. The most pronounced increases observed under drought conditions, when elevated AFB1 levels in maize-based feed led to increased AFM1 excretion in milk. Based on these datasets, AFM1 concentrations in both raw and processed milk in Hungary generally remain in the low-to-moderate range, with average values typically between 10 and 30 ng/L. In the Hungarian samples analysed by Farkas et al. (2022), raw milk showed a mean AFM1 concentration of 30.7 ± 24.7 ng/L, while heat-treated consumer milk had a mean of 18.0 ± 10.9 ng/L. In the same study, 9.4% of raw milk samples exceeded the EU regulatory limit of 50 ng/L, whereas only 0.5% of consumer milk samples were above this threshold.

Overall, the available evidence indicates that AFM1 contamination in Hungary does not follow a consistent increasing trend, but rather shows intermittent fluctuations driven primarily by climatic variability. Since the strengthening of the monitoring system after 2013, AFM1 levels in milk have generally remained below regulatory limits, and consumer exposure is considered low. Nevertheless, continuous surveillance remains essential due to the strong dependence of aflatoxin occurrence on weather conditions and feed quality.

Our Research and Findings

A study (Buzás et al., 2023) was conducted between 2022 and 2023 to monitor aflatoxin M1 (AFM1) concentrations and to estimate aflatoxin exposure in the adult population in Hungary. The study was published in the Journal of Food Composition and Analysis (Impact Factor: 4.6). The objective was to investigate seasonal variations in AFM1 levels in raw and drinking milk and to assess dietary exposure among Hungarian population.

A total of 474 milk samples, including raw milk (n = 278) and commercial milk (n = 196), were collected and analysed between September 2021 and November 2022. In parallel, 196 commercial milk samples — comprising pasteurized (n = 20), Extended Shelf Life (ESL, n = 94), and ultra-high temperature (UHT, n = 82) milk — were randomly purchased from supermarkets and retail outlets in Mosonmagyaróvár, located in north-western Hungary (Győr-Moson-Sopron County).

AFM1 concentrations were determined using an Enzyme-Linked Immunosorbent Assay (ELISA). Analytical determination was performed using the RIDASCREEN® Aflatoxin M1 ELISA Kit, a validated competitive enzyme immunoassay. The limit of detection (LOD) for AFM1 was 5 ng/L, with a reported selectivity of 100%. Milk samples were analysed in duplicate, and the absorbance values from the ELISA assay were processed using RIDASOFT© Win.net (Z9996FF) software to determine AFM1 concentrations. Results were expressed as mean ± standard deviation, median, and range (ng/L), along with the percentage of samples exceeding the limit of detection (LOD, 5 ng/L) and the EU maximum residue limit (MRL, 50 ng/L). Statistical analysis was performed using IBM® SPSS® Statistics (version 20, Chicago, IL, USA). Seasonal differences in AFM1 concentrations were evaluated using one-way analysis of variance (ANOVA), followed by Tukey's HSD post hoc test. A p-value of less than 0.05 was considered statistically significant. Dietary exposure and health risk assessment were evaluated by calculating the Average Daily Intake (ADI), Margin of Exposure (MOE), Hazard Index (HI), Cancer Potency (CP), and liver cancer risk (CR).

Results - Seasonal occurrence of AFM1 in raw milk samples

Between 2021 and 2022, a total of 278 raw milk samples and 196 drinking milk samples collected in Hungary were analysed for aflatoxin M1 (AFM1) contamination. AFM1 was detected in 68.7% of the raw milk samples, with an overall mean concentration of 30.7 ± 24.7 ng/L and values ranging from 5.0 to 173 ng/L. The highest contamination levels were observed during the autumn months (September–November), particularly in autumn 2022, when the average AFM1 concentration reached 39.5 ± 27.5 ng/L. During this season, 17.5% of the analysed raw milk samples exceeded the European Union maximum limit of 50 ng/L. In comparison, considerably lower concentrations were measured in spring and summer, with mean values of 19.6 ± 12.6 ng/L and 13.2 ± 5.6 ng/L, respectively, and no exceedances of the EU limit were detected during these periods. Statistical evaluation confirmed that AFM1 concentrations measured in autumn were significantly higher ($p < 0.05$) than those recorded in winter, spring, and summer seasons.

The analysis of drinking milk samples showed a similarly high occurrence of AFM1 contamination. Overall, 79.1% of the samples contained detectable AFM1 concentrations, ranging between 5.3 and 100.3 ng/L, with a mean concentration of 18.0 ± 10.9 ng/L. The highest contamination levels were again observed during autumn 2022, when almost all samples (97.4%) tested positive for AFM1, with an average concentration of 29.6 ± 13.5 ng/L. Despite the high frequency of contamination, only one sample exceeded the EU regulatory limit. In contrast, autumn 2021 showed substantially lower contamination, with only half of the drinking milk samples containing detectable AFM1 and an average concentration of 12.4 ± 11.1 ng/L.

The elevated AFM1 concentrations observed in autumn 2022 were most likely associated with the extreme climatic conditions experienced during the preceding summer. Hungary experienced unusually high temperatures and severe drought during 2022, conditions that favour the growth of aflatoxin-producing fungi in feed crops, particularly maize. Increased aflatoxin B1 (AFB1) contamination in feed subsequently results in elevated AFM1 levels in milk, as AFM1 is formed during the metabolic conversion of AFB1 in dairy animals. Seasonal differences may also be influenced by feeding practices, since during autumn and winter dairy cattle rely more heavily on stored feed materials such as silage and maize-based supplementary feed, which may contain higher concentrations of aflatoxins compared to fresh green fodder available during spring and summer.

Comparable seasonal trends have been reported in several neighbouring and European countries. Croatian monitoring studies identified the highest AFM1 concentrations during autumn, while surveys conducted in Albania and Serbia also demonstrated pronounced seasonal fluctuations in milk contamination levels. Particularly severe contamination events were reported in Serbia, where a substantial proportion of milk samples exceeded the EU regulatory limit during certain years. These findings support the conclusion that climatic conditions, especially heat and drought stress, play a major role in the occurrence and seasonal variation of AFM1 contamination in milk.

The exposure assessment showed seasonal variation in the average daily intake (ADI) of aflatoxin M1 (AFM1) from milk consumption in Hungary. The highest exposure levels were observed in autumn 2022, with ADI values reaching 0.085 ng/kg BW/day for drinking milk and 0.090 ng/kg BW/day for raw milk in adults with a body weight of 70 kg. Despite this increase, the estimated exposure levels remained below the average European ADI value reported by JECFA (0.11 ng/kg BW/day). Compared with international studies, the Hungarian exposure values were lower than those reported in countries such as Albania, Serbia, Iran, and Pakistan.

Health risk assessment parameters, including Hazard Index (HI), Margin of Exposure (MOE), and Liver Cancer Risk (LCR), indicated generally low health risks associated with AFM1 consumption. HI values remained below 1 in all seasons, suggesting no significant non-carcinogenic health concern. MOE values were mostly above the critical threshold of 10,000, except during autumn 2022, when elevated contamination temporarily reduced these values. Overall, the results indicate that AFM1 exposure through milk consumption does not currently represent a significant health risk for the adult population in Hungary.

Overall, AFM1 concentrations exceeded the European Union regulatory limit (50 ng/L) in 11.9% of raw milk samples and in only 0.5% of drinking milk samples. Although the calculated health risk assessment parameters (ADI, HI, MOE, and LCR) indicate that current consumer exposure to AFM1 in Hungary is generally low, the results highlight the importance of continuous monitoring and effective "farm-to-fork" control strategies. In the context of climate change and increasing frequency of hot and dry periods, regular surveillance of aflatoxins in feed and milk remains essential to minimise long-term food safety risks and maintain AFM1 concentrations below regulatory limits.

Conclusion and recommendation

The Visegrád countries share similar agroclimatic conditions and therefore have a similar risk profile regarding aflatoxin contamination. Favourable conditions for the proliferation of aflatoxin-producing Molds and toxin production are being created by extreme weather events, such as droughts and heat waves, which are becoming increasingly frequent due to climate change caused by global warming. The risk of aflatoxin in the V4 countries is therefore set to rise further in the future, which could also lead to significant economic and food safety challenges.

The V4 countries have robust agricultural and food industry trade, encompassing both raw materials and processed products. The EU has harmonised limits for aflatoxin, but it would be advisable in the future to establish a common, weather-based forecasting and warning system. This system would be capable of dynamically predicting increased aflatoxin risk using meteorological data. For instance, during extended periods of dry and hot weather, the system could automatically initiate enhanced food and feed control measures. Such a system could contribute to reducing health risks and economic losses.

Moreover, the development of new analytical methods within the framework of joint R&D projects among member states, as well as the conducting of aflatoxin ring trials, would result in the comparability of measurements performed by laboratories on identical samples. This would enable any methodological discrepancies to be identified, thereby improving measurement accuracy, strengthening quality assurance and promoting the development of uniform laboratory practices in the region.

Furthermore, researchers should focus on testing biocontrol solutions, specifically selecting and studying toxin-producing fungal strains capable of effectively suppressing aflatoxin-producing fungal species in both soil and plants.

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