MYCOBIOTA AND MYCOTOXINS IN FRESHLY HARVESTED AND STORED MAIZE


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Abstract: The incidence of mycobiota and mycotoxin levels were investigated in the freshly harvested maize kernel samples from October 2014 and in the samples of stored maize kernels from February 2015. Toxigenic fungal species (moulds) were isolated, cultivated and identified on agar plates according to standard mycological methods, while mycotoxins were detected by enzyme-linked immuno-sorbent assay (ELISA).

Mycological analyses of kernels showed the presence of toxigenic species from genera Aspergillus, Fusarium and Penicillium. Among the Aspergillus species, Aspergillus flavus was identified with higher incidence in the stored kernels (10.25%) than in freshly harvested kernels (3.67%) whereas A. parasiticus was the predominant species in the freshly harvested kernels (4.17%) compared to the stored kernels (0%). From the genus Fusarium three species were identified: F. graminearum, F. subglutinans and F. verticillioides, with the incidence of 1.08%, 8% and 25.75%, respectively in freshly harvested kernels and the incidence of 2.50%, 7.10% and 29.75%, respectively in the stored kernels. Species from genus Penicillium had higher incidence in freshly harvested kernels (14.25%) than in the stored kernels (9%).

In addition, tested samples of harvested and stored maize kernels were 100% positive with aflatoxin B₁ (AFB₁), deoxynivalenol (DON) and total fumonisins B₁, B₂ and B₃ (FBs). The mean levels of AFB₁, DON and FBs were 2.77 μg kg⁻¹, 117.83 μg kg⁻¹, and 3700.84 μg kg⁻¹, respectively in the freshly harvested kernels and a mean levels of 2.16 μg kg⁻¹, 2034.40 μg kg⁻¹, and 5976.50 μg kg⁻¹, respectively in the stored maize kernels.

In the freshly harvested maize kernel samples, statistically significant (P ≤ 0.05) positive correlations of kernel moisture content with the incidence of Penicillium spp. (r = 0.47), and levels of AFB₁ (r = 0.46) and FBs (r = 0.47), and between the incidence of Penicillium spp. and level of AFB₁ (r = 0.53) were established. In the stored maize kernel samples, statistically significant (P ≤ 0.05) positive correlations were found between the incidence of F. subglutinans and level of FBs (r = 0.50) and between levels AFB₁ and FBs (r = 0.52). A highly significant
(P ≤ 0.01) positive correlation was established between the incidence of *F. verticillioides* and level of FBs (r = 0.64) in freshly harvested maize kernel samples.

These results indicate that the incidence of toxigenic fungi and levels of mycotoxins, in particular DON and FBs, were higher in the stored maize kernel samples than in freshly harvested maize kernels. Therefore, to prevent the development of toxigenic fungi and mycotoxins accumulation in post-harvest period it is necessary to thoroughly dry maize and keep it in hygienic food storages.

**Key words**: mycobiota, mycotoxins, harvest, storage, maize

**Introduction**

Maize is cereal crop used for human and animal nutrition. According to data of *Statistical Yearbook of Serbia* (2012), maize has been grown on about 1.2 million hectares with an average yield of 5.4 t ha⁻¹ and with a production of 6.5 million tons in Serbia in 2011.

The contamination of maize with fungi (moulds) and their secondary metabolites (mycotoxins) represents a serious hazard to humans and animals. The most important mycotoxins in maize kernels are aflatoxins produced by *Aspergillus flavus* and *A. parasiticus*. Equally important mycotoxins in maize kernels are fumonisins produced mostly by *Fusarium verticillioides* and *F. proliferatum*, as well as deoxynivalenol produced by *F. graminearum* (Chulze, 2010; Pereyra et al., 2011).

Climatic conditions and maize growing on large areas in Serbia are suitable for development of numerous toxigenic species, resulting with frequent animal feed contamination by toxic products of fungi – mycotoxins. Development of toxigenic fungi and bio-synthesis of mycotoxins most often depend on ample precipitation and low temperatures at the end of summer or beginning of autumn during sensitive phenophase of maize growing (Krnjaja et al., 2009). Mould growth and mycotoxin contamination can occur in the field pre-harvest and later during storage condition in the post-harvest period. The higher moisture and temperature are abiotic factors which positively influence on fungal growth and mycotoxin accumulation (Niaz et al., 2011; Kocasari et al., 2013). Production of farm animals, poultry and swine, in particular, requires a large amount of cereal grain. For this reason, most of the grain is stored until utilized. Storage conditions are determined by abiotic and biotic factors including microorganisms, insects, mites, rodents and birds (Santin et al., 2005).

Consumption of mycotoxin contaminated diet may cause acute and chronic toxicity in humans and animals. Among farm animals, swine are considered to be
the most sensitive species to mycotoxins. Aflatoxin B₁ (AFB₁) is main hepatotoxin that causes various pathological effects on organs and tissues. Deoxynivalenol (DON) causes feed refusal, emesis, anaemia, haemorrhage, immunosuppression and neurotoxic effects. Fumonisins B₁, B₂ and B₃ (FBs) cause leukoencephalomalacia and porcine pulmonary edema (Biagi, 2009; Berardo, 2011; Pereyra et al., 2011).

Unfortunately, there are no direct measures for prevention of fungal infection and mycotoxin contamination. However, unfavourable conditions for development of toxigenic fungi could be provided by implementation of appropriate agricultural practices as preventive measures in the field. Pre-harvesting control strategies primarily consist of crop practices designed to reduce the fungal development and mycotoxin accumulation and the utilization of genetically resistant hybrids (Blandino et al., 2008). Post-harvest control strategy includes the practices directed at reducing mycotoxin levels, mycotoxin risk assessment in crop products and controlling its use through regulation. Monitoring of fungal and mycotoxin contamination in crops and products can be implemented in pre-harvest and post-harvest period (Dohlman, 2003). Reduction moisture in grains to moisture level of 13% or below is also very important post-harvest control measure (Lutfy et al., 2008).

The aim of this research was to determine the incidence of toxigenic fungal species, levels of mycotoxins (AFB₁, DON and FBs) and to establish the signification of correlation coefficients between investigated variables in freshly harvested and stored maize kernel samples, used for animal feed.

**Material and Methods**

During the harvest time in October of 2014 and storage time in February of 2015, 20 and 20 maize kernel samples intended for farm animals feeding (pigs, sheep and poultry) were randomly collected from parcels for agricultural production and warehouse with natural-air-drying conditions in the Institute for Animal Husbandry, Belgrade. The samples were collected according to the Commission Regulation (EC) No 401/2006 (European Commission, 2006). All samples were kept at 4ºC in the refrigerator before further analysis. Moisture content of milled maize kernels was determined using a moisture analyzer (OHAUS MB35, USA).

For the mycological analysis, maize kernels were disinfected in 1% sodium hypochlorite (NaOCl) for 2-3 minutes, and rinsed twice in distilled water. A total of 50 maize kernels per each sample were distributed in Petri dishes (5 kernels per Petri dish) with water-1.8% salt agar (18g NaCl per 1 litre of agar medium) (Krnjaja et al., 2015). Plates were incubated for 14 days at 20ºC with alternating light and darkness. Potential toxigenic fungi were identified according to the fungal
key of Singh et al. (1991). The incidence of potential toxigenic fungal species was calculated according to Lević et al. (2012).

For the mycotoxicological analysis, the maize kernel samples were ground to a fine powder with an analytical mill (IKA A11, Germany). The levels of AFB₁, DON and FBs were detected using the competitive ELISA method according to the manufacturer's instructions Celer Tecna® ELISA kits. Absorbance was determined at a wavelength of 450 nm on an ELISA plate reader spectrophotometer (Biotek EL x 800TM, USA). The limit of detection for AFB₁, DON and FBs were 1 μg kg⁻¹, 40 μg kg⁻¹ and 750 μg kg⁻¹, respectively.

The correlation between individual values for moisture content of maize kernels, the incidence of toxigenic fungal species and the levels of AFB₁, DON and FBs was determined using the Pearson correlation coefficient.

Results

The average moisture content for freshly harvested maize kernel samples was 15.20% (range 14.28 – 16.05%) and for stored maize kernel samples was 14.04% (range 13.41 – 14.92%).

In mycological analyses, toxigenic fungi from genera Aspergillus, Fusarium and Penicillium were identified. Fungal species identified in both, freshly harvested and stored maize kernels, were A. flavus, F. graminearum, F. subglutinans, F. verticillioides and Penicillium spp., with the exception of the species A. parasiticus which was identified only in the freshly harvested maize kernels. The most frequent fungal species was F. verticillioides, with an average incidence of 25.75% (range 10 - 43%) in freshly harvested and 29.75% (range 17 – 50%) in stored maize kernels. Considering the average values, the incidence of A. flavus was higher in stored (10.25%) than in freshly harvested maize kernels (3.67%). The species A. parasiticus was identified only in freshly harvested kernels with presence of 4.17% on average. The presence of F. graminearum was higher in stored (on average 2.50%) than in freshly harvested kernels (on average 1.08%), while the incidence of F. subglutinans and Penicillium spp. was higher in freshly harvested (on average 8% and 14.25% respectively) than in stored maize kernels (on average 7.10% and 9% respectively) (Table 1).

In the mycotoxicological analyses, freshly harvested and stored maize kernels samples were 100% positive for the presence of all tested mycotoxins. In the freshly harvested samples the mean levels of AFB₁, DON and FBs were 2.77 μg kg⁻¹ (range 2.31 – 3.34 μg kg⁻¹), 117.83 μg kg⁻¹ (range 42 – 238 μg kg⁻¹) and 3700.84 μg kg⁻¹ (range 1519 – 9780 μg kg⁻¹), respectively (Table 2). In the stored maize samples the mean levels of AFB₁, DON and FBs were 2.16 μg kg⁻¹ (range 1.03 – 4.11 μg kg⁻¹), 2034.40 μg kg⁻¹ (range 380 – 10684 μg kg⁻¹) and 5976.50 μg kg⁻¹ (range 760 – 35760 μg kg⁻¹), respectively (Table 3).
Table 1. Incidence of fungal species in freshly harvested and stored maize kernel samples

<table>
<thead>
<tr>
<th>Fungal species</th>
<th>Freshly harvested kernel</th>
<th>Stored kernel</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Incidence (%)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Average</td>
<td>Range</td>
</tr>
<tr>
<td>Aspergillus flavus</td>
<td>3.67</td>
<td>0-10</td>
</tr>
<tr>
<td>Aspergillus parasiticus</td>
<td>4.17</td>
<td>0-17</td>
</tr>
<tr>
<td>Fusarium graminearum</td>
<td>0.08</td>
<td>0-7</td>
</tr>
<tr>
<td>F. subglutinans</td>
<td>8</td>
<td>0-20</td>
</tr>
<tr>
<td>F. verticillioides</td>
<td>25.75</td>
<td>10-43</td>
</tr>
<tr>
<td>Penicillium spp.</td>
<td>14.25</td>
<td>0-27</td>
</tr>
</tbody>
</table>

Table 2. Levels of aflatoxin B1 (AFB1), deoxynivalenol (DON) and total fumonisins (FBs) in freshly harvested maize kernel samples

<table>
<thead>
<tr>
<th>Item</th>
<th>Freshly harvested kernel</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>AFB1</td>
<td>DON</td>
</tr>
<tr>
<td>Sample size&lt;sup&gt;a&lt;/sup&gt;</td>
<td>20/20</td>
<td>20/20</td>
</tr>
<tr>
<td>Incidence (%)</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Range (μg kg&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>2.31-3.34</td>
<td>42-238</td>
</tr>
<tr>
<td>Mean&lt;sup&gt;b&lt;/sup&gt; (μg kg&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>2.77</td>
<td>117.83</td>
</tr>
</tbody>
</table>

<sup>a</sup> Number of positive samples/Number of total samples  
<sup>b</sup> Mean level in positive samples

Table 3. Levels of aflatoxin B1 (AFB1), deoxynivalenol (DON) and total fumonisins (FBs) in stored maize kernel samples

<table>
<thead>
<tr>
<th>Item</th>
<th>Stored kernel</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>AFB1</td>
<td>DON</td>
</tr>
<tr>
<td>Sample size&lt;sup&gt;a&lt;/sup&gt;</td>
<td>20/20</td>
<td>20/20</td>
</tr>
<tr>
<td>Incidence (%)</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Range (μg kg&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>1.03-4.11</td>
<td>380-10684</td>
</tr>
<tr>
<td>Mean&lt;sup&gt;b&lt;/sup&gt; (μg kg&lt;sup&gt;-1&lt;/sup&gt;)</td>
<td>2.16</td>
<td>2034.40</td>
</tr>
</tbody>
</table>

<sup>a</sup> Number of positive samples/Number of total samples  
<sup>b</sup> Mean level in positive samples

Statistical analyses and Pearson’s correlation coefficient, established highly significant positive correlation (P ≤ 0.01) between the incidence of *F. verticillioides* and level of FBs (r = 0.64) in the freshly harvested kernel maize samples. There was also significant positive correlation (P ≤ 0.05) of kernel moisture content with levels of AFB1 (r = 0.46) and FBs (r = 0.47) and between the incidence of *Penicillium* spp. and level of AFB1 (r = 0.53). Likewise, the positive correlation, but not statistically significant, was found between kernel moisture content and level of DON (r = 0.27), then between levels of AFB1 and FBs (r = 0.31), between the incidence of *F. subglutinans* and level of FBs (r = 0.30), and between the incidence of *A. flavus* and AFB1 (r = 0.20). It was only established the
statistically insignificant negative correlation between the incidence of *A. parasiticus* and level of AFB$_1$ ($r = -0.06$) in the freshly harvested maize kernels. Considering correlation coefficients in the stored maize kernel samples, statistically significant positive correlation ($P \leq 0.05$) was also found between levels of AFB$_1$ and FBs ($r = 0.52$), then between the incidence of *F. subglutinans* and level of FBs ($r = 0.50$) and between the incidence of *A. flavus* and AFB$_1$ ($r = 0.34$). There were also positive correlations but not significant between the incidence of *F. verticillioides* and level of FBs ($r = 0.31$), then positive correlation of kernel moisture content with levels of FBs ($r = 0.26$), AFB$_1$ ($r = 0.17$) and DON ($r = 0.03$). The negative, but not significant, correlation was found only between the incidence of *F. graminearum* and level of DON ($r = -0.30$) in the stored maize kernel samples (data not presented).

**Discussion**

In this study the incidence of mycobiota and levels of mycotoxins in the samples of freshly harvested and stored maize kernels were researched. The contamination of cereals with toxigenic fungi under favourable climatic and storage conditions may lead to mycotoxin accumulation to injurious levels for farm animals and human health. The production of mycotoxins is consequence of increased presence of toxigenic fungi. Therefore, determination of fungal species at the right time is very important step to reduce the detrimental effects of mycotoxin problems.

All tested samples of freshly harvested and stored maize kernels were positive for the presence of mycotoxins AFB$_1$, DON and FBs. In samples of freshly harvested maize kernels, mean levels of mycotoxins did not exceed the maximum permitted limits for unprocessed maize prescribed by the regulations of the Republic of Serbia (*Sl. glasnik*, 2014), but in some samples FBs level was above the maximum permitted limit (4000 μg kg$^{-1}$), with a maximum level of 9780 μg kg$^{-1}$ (Table 2). In samples of stored maize kernels, mean level of DON was above the maximum permitted limit (1750 μg kg$^{-1}$) with a maximum level of 10684 μg kg$^{-1}$, as well as certain samples with FBs levels above the maximum permitted limit – maximum level of 35760 μg kg$^{-1}$ (Table 3). The incidence of DON producer, *F. graminearum*, and FBs producers, *F. subglutinans* and *F. verticillioides*, was higher in the stored than in the freshly harvested maize kernels (Table 1). These results can be explained by the favourable climate conditions during the maize harvest in 2014 and during the winter period in 2015. Namely, according to data of the Republic Hydro-meteorological Service of Serbia for 2014, heavy total rainfall was recorded in September (126 mm) with a mean daily temperature of 18.3°C before maize harvest. For this reason the average moisture content of harvested maize kernels was high (> 15%). After harvest, the maize was stored in a warehouse and naturally dried without conditions control (temperature and
humidity). Mild winter during 2015 and uncontrolled conditions of temperature and relative humidity in the warehouse caused the intensive development of moulds and increased mycotoxins content, especially of DON in samples of stored kernels.

Similarly to this, in earlier mycotoxicological studies in Serbia, Krnjaja et al. (2013a) have found in maize kernels harvested in 2012 the mean level of AFB$_1$ in 100% of tested samples exceeding the maximum permitted limit (5 μg kg$^{-1}$; Sl. glasnik, 2014) in unprocessed maize, with incidence of A. flavus of 36.69%. These authors also concluded that the mean levels of DON and FBs have not exceeded the maximum permitted limits. It has been assumed that the drought in 2012 had also been the reason for the occurrence of aflatoxigenic fungi and high level of AFB$_1$ up to >40 μg kg$^{-1}$. In addition, in the stored maize samples collected from October 2011 to October 2012 with an average kernel moisture content of 11.02% Krnjaja et al. (2013b) have detected AFB$_1$, DON and FBs with the mean levels of 1.39 μg kg$^{-1}$, 128.17 μg kg$^{-1}$ and 1610.83 μg kg$^{-1}$, respectively. Compared to year 2014, in 2011 less rainfall was recorded before the maize harvest in September (47.7 mm) with a mean daily temperature of 17.2°C. Such climatic conditions before harvesting and especially kernel moisture content during the period of maize storage were not suitable for intensive development of toxigenic fungi and thus for the increased production of mycotoxins.

In total 26 maize samples from Turkey (n=19) and USA (n=7) collected between April 2002 and 2003, the mean aflatoxin and fumonisins levels were higher in samples from Turkey (10.94 μg kg$^{-1}$ and 88240 μg kg$^{-1}$, respectively) than in samples from USA (0.78 μg kg$^{-1}$ and 74150 μg kg$^{-1}$, respectively) (Oruc et al., 2006). By examining a total of 82 consignments of French and Argentinean maize as raw, imported in the United Kingdom between 2004 and 2007, Scudamore and Patel (2009) have detected the maximum level of 444 μg kg$^{-1}$ for DON and 5002 μg kg$^{-1}$ for FBs. These authors have found clear differences in the levels of mycotoxins between harvests and geographic regions. Maize from Argentine contained lower levels of DON and higher levels of FBs than maize from France, although the level of FBs up to 2000 μg kg$^{-1}$ or more were present in samples taken from both regions. Likewise, levels of fumonisins were higher in 2004 in Argentina and in 2006 in France due to exceptionally hot and dry summer and dry period before harvest. In the USA, Dowd and Johnson (2010) over a 4-year period (2005-2008) conducted mycotoxicological studies of popcorn samples and have rarely detected AFB$_1$, but FBs and DON were present in all years, with mean levels in fields up to 1700 μg kg$^{-1}$ (sample max. 2770 μg kg$^{-1}$) and 1900 μg kg$^{-1}$ (sample max. 2660 μg kg$^{-1}$). These authors have concluded that the damage from insects is the main cause of higher levels of FBs in relation to the DON levels, while higher levels of DON are caused by higher rainfall and lower temperatures during the maize ripening. In mycotoxicological analysis of 2258 maize samples collected over a 3-year period (2006-2008) from 93 storage centres in Italy, Berardo et al.
have found a high level of FBs, with the highest mean level in 2006 (10900 μg kg⁻¹) and lowest in 2008 (4800 μg kg⁻¹). These authors have assumed that climatic factors in conjunction with the specific growing area, played an important role in the accumulation of FBs in maize. In maize samples collected from July through August 2011, from 15 swine farms in the Beijing region in China, Li et al. (2014) have reported the natural occurrence of AFB₁ and DON with the highest levels of 58.9 μg kg⁻¹ (on average 6 μg kg⁻¹) and 2130 μg kg⁻¹ (on average 1091 μg kg⁻¹), respectively. Similar to the above mentioned data our results confirm high maximum levels of FBs in both harvested and stored maize kernels and high maximum level of DON in stored maize kernels.

After examining the correlation coefficients, in most variables from both tested groups in this research, harvested and stored maize samples, positive correlations were found, particularly emphasizing the positive correlation between the incidence of F. verticillioides and F. subglutinans with FBs level, and the incidence of A. flavus and AFB₁ level, then positive correlation between AFB₁ and FBs levels and between kernel moisture content and DON level. These results are similar to studies of Kimanya et al. (2008), Sun et al. (2011), Berardo et al. (2011) and Krnjaja et al. (2013a).

In this study, in tested harvested and stored maize kernel samples, the presence of potentially toxigenic fungi and mycotoxins AFB₁, DON and FBs was recorded. Because of favourable conditions for the growth and development of toxigenic fungi and mycotoxin contamination of maize kernels, it is of utmost importance to implement the preventive measures to reduce the risk of these contaminants in Serbia, especially in years when weather conditions are suitable for their development. Preventive measures, such as fast drying of maize for the medium- and long-term storage in hygiene maintained warehouses, without the presence of insects and microorganisms, and proper regulation of the moisture content of kernels, could significantly reduce the mycotoxins contamination of maize grains (Bruns, 2003). Good ventilation of warehouse has also been one of the most important preventive measures for the reduction of mycotoxins production (Jakic-Dimic et al., 2011).

Conclusion

Fungal and mycotoxin contamination of maize has been increasing worldwide, as a result of climate change. Some mycotoxins could be synthesized in maize before harvest but their level may increase after harvest during the storage period and further in the food chain.

Based on the obtained results it can be concluded that the potentially toxigenic species of fungi from the genera Aspergillus, Fusarium and Penicillium were present in the harvested and stored maize kernel samples. Species of the genera Aspergillus and Fusarium had higher incidence in stored samples compared
to freshly harvested maize. Also, in both groups of maize samples, AFB₁, DON and FBs were detected, but in the samples of stored maize kernels, the mean levels of DON and FBs exceeded the maximum permitted levels for unprocessed maize as stipulated by Serbian Regulation. For this reason, constant supervision and monitoring of mycotoxins occurrence in maize pre- and post-harvest and application of preventive measures are very important to reduce risks to human and animal health.

Acknowledgment

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Mikobiota i mikotoksini u sveže požnjevenom i uskladištenom zrnu kukuruza


Rezime

U radu je proučavana učestalost mikobiota i sadržaj mikotoksina u uzorcima zrna kukuruza sakupljenih tokom berbe u oktobru 2014. godine i u uzorcima uskladištenog zrna sakupljenih u februaru 2015. godine. Toksigene vrste gljiva (plesni) su izolovane, odgajene i identifikovane na hranljivoj podlozi prema standardnim mikološkim metodama, dok je sadržaj mikotoksina detektovan primenom imunoadsorpcione enzimske metode (ELISA).

Mikološkim analizama zrna kukuruza ustanovljeno je prisustvo toksigenih vrsta iz rodova Aspergillus, Fusarium and Penicillium. Među Aspergillus vrstama, Aspergillus flavus je identifikovana u većem procentu u uzorcima uskladištenog zrna (10,25%) nego u uzorcima sveže požnjevenog zrna (3,67%), a A. parasiticus bila je predominantna vrsta (4,17%) u uzorcima sveže požnjevenog u odnosu na uskladišteno zrno kukuruza (0%). Tri vrste roda Fusarium su identifikovane sa učestalošću od 1,08% (F. graminearum), 8% (F. subglutinans) i 25,75% (F. verticillioides) u požnjevenom zrnu, i sa učestalošću od 2,50% (F. graminearum), 7,10% (F. subglutinans) i 29,75% (F. verticillioides) u uskladištenom zrnu kukuruza. Vrste iz roda Penicillium imale su veću učestalost u uzorcima požnjevenog (14,25%) nego u uzorcima uskladištenog zrna (9%).

Ispitivani uzorci sveže požnjevenog i uskladištenog zrna bili su 100% pozitivni sa aflatoxinsom B₁ (AFB₁), deoksinivalenolom (DON) i ukupnim
fumonizinima \( \text{FB}_1 \), \( \text{FB}_2 \) i \( \text{FB}_3 \) (FBs). Prosječne koncentracije ovih toksina su iznosile 2,77 μg kg\(^{-1}\) (AFB\(_1\)), 117,83 μg kg\(^{-1}\) (DON) i 3700,84 μg kg\(^{-1}\) (FBs) u uzorcima sveže požnjevenog zrna i 2,16 μg kg\(^{-1}\) (AFB\(_1\)), 2034,40 μg kg\(^{-1}\) (DON), i 5976,50 μg kg\(^{-1}\) (FBs) u uzorcima usklađenog zrna.

Statistički značajne \( (P \leq 0.05) \) pozitivne korelacije ustanovljene su između sadržaja vlage zrna sa učestalosti \textit{Penicillium} spp. \( (r = 0.47) \) i koncentracijama \( \text{AFB}_1 \) \( (r = 0.46) \) i FBs \( (r = 0.47) \), kao i između učestalosti \textit{Penicillium} spp. i koncentracije \( \text{AFB}_1 \) \( (r = 0.53) \). U uzorcima usklađenog zrna, statistički značajne \( (P \leq 0.05) \) pozitivne korelacije ustanovljene su između učestalosti \textit{F. subglutinans} i koncentracije FBs \( (r = 0.50) \) i između koncentracija \( \text{AFB}_1 \) i FBs \( (r = 0.52) \). Statistički veoma značajna \( (P \leq 0.01) \) pozitivna korelacija ustanovljena je između učestalosti \textit{F. verticillioides} i koncentracije FBs \( (r = 0.64) \) u uzorcima sveže požnjevenog zrna kukuruza.

Rezultati ovih istraživanja ukazuju da su učestalosti toksigenih vrsta gljiva i koncentracije mikotoksina, posebno DON i FBs, bile više u uzorcima usklađenog zrna nego u uzorcima sveže požnjevenog zrna. Zbog toga, da bi se sprečio razvoj toksigenih gljiva i akumulacija mikotoksina u postžetvenom periodu neophodno je kukuruz dobro osušiti i čuvati u higijensko ispravnim skladištima.

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