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MYCOTOXINS IN FEED FOR PIGS AND POULTRY

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Abstract: Mycological and mycotoxicological analysis of many feedingstuffs and complete mashes for different animal species and categories have showen that the feed safety problem has been present for years. It is approved that mouldy feed can contain various mycotoxins, but among them in our conditions the most investigated are aflatoxins, ochratoxins, zearalenone and some trichotecenes. Based on laboratory feed inspection of samples sent to be analysed in the Institute of Veterinary Medicine of Serbia in Belgrade it was concluded that fungi and mycotoxins contamination are common. Primarly presence of zearalenone, ochratoxine and aflatoxin was proven. Although these levels usually do not exceed limits layed by actual legislation, having on mind cumulative effects and possibal chronical exposure of animals to their harmful influence, appropriate and competent approach is necessary. In fact, even when direct loses, as concequence of high mortality are not present, indirect loses due to drop of animal performances and production and occurence of secondary infections must not be neglected.

Key words*:* feed, fungi, mycotoxins

Introduction

Mycological and mycotoxicological analysis of feed used in the nutrition of various categories of poultry and swine showed that feed hygiene and safety had been a topical issue for years. Mycotoxins are toxic secondary metabolites of fungi, which enter organisms of animals usually over intake of contaminated feed. Through such feed, that contains mycotoxins, in the animal organism appear intoxication, so called mycotoxicoses, that can induce a wide scale of adverse effects *(Uraguchi and Jamazaki, 1978)*. Damages to livestock breeding, which result from mycotoxicoses can be large. They are manifested in the form of direct losses, due to high mortality, or more often they occur as indirect losses due to falling production and reproductive ability of animals. A special problem is that the contaminated feed contains a variety of mycotoxins in different quantities, which also express the difference in the harmful effects.

So far, several hundred mycotoxins are discovered among which a small number is considered to be detrimental *(Riley, 1998)* and only 20% to 30% according to the frequency of occurrence and harmful effects have a medical, nutritional, environmental and economic importance. Mould and mycotoxin contamination is a worldwide problem because of the 25% to 35% world cereals are contaminated with fungi *(Ueno, 1983)*. Growth and development of fungi in animal feed causes the change of many properties, reduces the nutritional value of feed, up to 50% reduces content of dry matter *(Lazzari, 1993)* and energy value of feed by lowering fat and carbo hydrates content and reduces the metabolic energy *(Bartov, 1982)*. In this way, the costs of feeding are significantly increased. In the period of prevalence of unfavorable external conditions production of fungi secondary metabolites, ie mycotoxins, is induced. The most common moulds are from *Aspergillus, Penicillium*, *Rhizopus* and *Fusarium* genera, which produce alfatoxins, ochratoxins, zearalenone and trichotecens. According to the primary effect in the organisms, mycotoxins are divided into hepatotoxic, nephrotoxic, neurotoxic and cytotoxic *(Sinovec et al., 2000)*. According to the biological effects they may be carcinogenic, mutagens, teratogens, immuno modulators and inhibitors of protein synthesis.

Aflatoxins are hepatotoxins which are products of the fungi from genus *Aspergillus*. They are metabolites of *Aspergillus flavus* and *Aspergillus parasiticus* only *(Diener and Daviš, 1966)*. *Aspergillus flavus* is a mould which is very widespread in nature, and can be found in corn, wheat, barley, oat, rice, etc. Fungi produce aflatoxins in the presence of higher moisture, temperature and adequate substratum. Aflatoxicosis is manifested by various patomorphological changes in the liver, kidneys and nervous tissue. The adverse effects are manifested on the production results, especially in the broiler breeding, then in swine production, as well as other types of livestock production.

Ochratoxin is the product of fungi in the field and during storage. Fungi of Fungi species from *Penicillium* and *Aspregillus* genera produce it *(Wullie, Morechause, 1972)*. Ochratoxin is slowly apsorbed from the digestive tract. After the resorption most of ochratoxin is present in the kidneys, liver and muscles. According to toxicity ochratoxin is one of the most toxic mycotoxins *(Uragnchi and Jamazaki, 1978)*. Pigs are the most sensitive animals, with the occurrence of nephropathy *(Krogh, 1978)*. Poultry is less sensitive. In laying hans the fall of egg production and the appearance of nephropathy can be noted *(Nedeljkovic et al., 2003)*.

Zearalenone is a toxic product of *Fusarium* fungi that contaminate more grain in the field, but the growth of mould and synthesis of toxins also continue in warehouses. After oral ingestion zearalenone resorption is very fast and it is transported to the liver. Pigs are the most sensitive species of animals *(Ozegovic, 1995)*, while ruminants and poultry, especially chickens react less sensitive.

T-2 toxin is a toxic product of fungi genus *Fusarium*. After ingestion it is very quickly resorbed in front parties of digestive tract. After 3 to 4 hours toxin is in the most organs: liver, kidneys, muscles. It is believed that trichotecenes are significantly more toxic than the other fungi metabolites. The most sensitive animals are pigs and poultry, while the ruminants are the least sensitive because of the effect of their rumen microflora. T-2 toxin has a strong immunosuppressive effect, as well as most of other trichotecens. T-2 toxin damages different tissues and organs *(Sinovec and Jovanovic, 1993)*.

The paper presents results of mycotoxicological analyses of different samples of feed used in nutrition of poultry and pigs.

Material and Methods

In order to determine levels of contamination of feedingstuffs and complete mashes used in animal nutrition mycotoxicological analysis of collected samples in the period from 2007 until 2008 were done. A total of 107 samples of feed for piglets, 55 mixtures for fattening pigs, 46 feed samples for pigs in reproduction, 68 samples of broiler feed, 107 samples of mashes for layers and 88 samples of chicken feed.

In all samples presence of mycotoxins was detected by ELISA method which was based on antigen-antybody reaction. Into the wells of microtitar plate, according to directions of commercial kit (R-Biopharm®) standards and samples were introduced. To make reaction visible chromogen had to be supplemented, which in contact with enzymes became bly, but after addition of stop reagent it became yellow. At the end adsorbance was measured fotometricly at the ELISA reader and, concidering the standard curve, calculation of mycotoxins content in the sample was done precisely.

All results were compared and interpreted regarding national Directive on maximal amounts of harmful materials in feed *(Službeni list SFRJ, 1990)*.

Results and Discussion

Among 263 samples of poultry feed in the 51.3% the presence of mycotoxins was found. In the feed for broilers ochratoxin was present in 43.3% of samples within the limits permitted by Regulation (1 mg/kg). The results of analysis for 88 mixtures for chickens and 107 mixtures for layers showed that 46% were contaminated by this toxin, but as the limit is significantly lower (0.25 mg / kg) for layers 10% of these samples exceeded the prescribed level. In 64.6% of samples of poultry feed was found the presence of zearalenone, but because of tolerance to this kind of toxins and the allowed limit of even 100 mg/kg, there were

no clinical or legal consequences. Aflatoxin and T-2 toxin in feed samples for poultry were not exceeded maximum levels specified in the regulations.

The analysis of the results of mycotoxicological testing of 107 samples for feeding of piglets, 55 mixtures for fattening pigs and 46 samples of mashes for pigs in reproduction it was noted that 26.4% of all contained aflatoxin in the amount higher than the limits prescribed by the Directive (0.02 mg/kg). The same percentage of deviation above allowable limits was found for ochratoxin content, which is limited to 0.1 mg/kg for piglets, and for other categories of pigs 0.2 mg/kg feed, while the maximal detected amount was 0.32 mg/kg feed. In the feed for these animals was proven the presence of zearalenone in 31.3% of samples in the amount above the level stated in the Directive (for piglets and pigs up to 50kg 0.5, and for other categories 1.00 mg/kg feed). T-2 toxin was found in 5.4% of analyzed samples.

Table 1. Presence of mycotoxins above prescribed limits

	Ochratoxin	Zearalenone	Aflatoxin	T-2 toxin
Type of feed	Frequency of mycotoxins (%)			
Poultry feed	10 %	-	-	-
Swine	26.4 %	31.3 %	26.4 %	-

In feed for pigs and poultry, there is always a certain number of microorganisms of different origin. The presence and a number of bacteria and fungi (*Mucor, Rhizopus, Penicillium*, etc.) are directly related to the composition of produced mixtures. Composing the meal for animals the largest part, over 50%, makes corn and other cereals. Therefore, in the territory of our country the most frequently is contaminated feed for the pigs and corn, which can even contain over 10 mg/kg zearalenone.

Food and feed safety and hygiene are a significant problem, and great attention is directed towards diseases that are closely related to different mycotoxicoses. Mycotoxins cause a whole range of disorders in the body of animals, ranging from biochemical changes, through the functional and morphological damages of different tissues and organs, to the appearance of clinical signs of mycotoxicoses with even possible lethal outcome.

Small amounts of mycotoxins in feed can provoke latent daily changes in the intestines: the reduction of resorption of amino acids and vitamins, while at the same time stimulate the creation of free radicals in the intestinal tract. They are absorbed in the intestines and accumulate in the various tissues. Have immunosuppressive effects, leading to damage of receptor cells and the changes in the structure of DNA and RNK molecules.

Improvement of the quality of grain storage and production and conditions in warehouses for final products (lower humidity, good ventilation etc.) it is

possible to reduce the synthesis of mycotoxins, which is one of the most important aspects of prevention of this problem.

Conclusion

Mycotoxicological analyses of samples of feed for poultry and pigs have shown that the presence of mycotoxins, despite the attention that was paid during last years, is still very actual problem.

Contamination is especially frequently present in feed for pigs, and in the almost third of analyzed samples it exceeds maximum allowable limits prescribed by Directive on maximal amounts of harmful materials in feed *(Službeni list SFRJ, 1990)*.

However, in the case of lower levels of mycotoxins still remains the risk of rigid interpretation of the regulations, as it is well known that the use of feed with low content of mycotoxins in the longer time period shows similar effects as short-term use of feed with higher amount of mycotoxins. The problem is getting more complicated as the interaction of mycotoxins present in feed increases harmful effects. Therefore, the assessment of utilization of feed should still depend on the institutions and experts that this problem is one of the special scientific and professional activities.

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Prisustvo mikotoksina u smešama za ishranu svinja i živine

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Rezime

Mikotoksikološkim ispitivanjem uzoraka hrane za živinu i svinje utvrđeno je da je prisustvo mikotoksina veoma aktuelan problem. Pokazalo se da je naročito učestala kontaminacija mikotoksinima prisutna u hrani za svinje, i to u skoro trećini analiziranih uzoraka prevazlazeći maksimalno dozvoljene granice Pravilnikom o maksimalnim količinama štetnih materija i sastojaka u stočnoj hrani *(Službeni list SFRJ, 1990)*. Imajući u vidu kumulativni efekat i moguću hroničnu izloženost životinja štetnom dejstvu mikotoksina i u slučaju nižih koncentracija potrebna je odgovarajuća stručna kontrola. I onda kada se ne manifestuju direktni

gubici kao posledica uginjavanja životinja, ne smeju se zanemariti indirektne štete usled pada proizvodnje i pojave sekundarnih oboljenja.

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