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HARMFUL ORGANISMS OF GRAIN AS POTENTIAL RISKS TO HUMAN HEALTH

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Abstract

One of the main imperatives of agriculture and processing industry is the production of safe food of high nutritional quality free of biological, physical and chemical contaminants. Cereals are an essential part of human nutrition. Microorganism represents permanent microflora elements of cereals and fruits. The most frequent moulds are *Aspergillus*, *Penicillium*, *Fusarium*, *Claviceps* and *Alternaria*. Some of them biosynthesizes and excrete mycotoxins (aflatoxins, ochratoxins, sterigmatocystin, zearalenone, fumonisin, deoxynivalenol, ergot alkaloids). Data testify that about 25% of the total world production of grain is contaminated with at least one mycotoxin. Among the raw materials that are usually contaminated with mycotoxins are cereals, maize, oilseeds, dry and stone fruits. Mycotoxins in humans and animals cause mycotoxicosis and manifest mutagenic, teratogenic, embryo-toxic, hepatogenic, nephrotoxic, dermatogenic and carcinogenic effect. Stored products are also attacked by storage pests. The most important insect storage pests are *Sytophilus granaries*, *Rhyzopertha dominica*, *Tribolium* sp., *Pyralis farinalis*, *Sitotriga cerealella*, *Plodia interpuctella*, *Ephestia kuehniella* and *Tinea granella*. Contamination by insects, their metabolites and excrements reduces the quality of the food and makes it unsuitable for consumption. Many insect species are hosts and vectors of pathogens of man and/or animals. Carcinogenic and teratogenic substances are produced only by *Tribolium* sp. Professional exposure to the infested dust of grains is associated with changes in the type of allergic conjunctivitis, dermatitis or asthma. From rodents as storage pests and vectors of infectious diseases emphasize *Ratus norvegicus*, *Mus musculus* and *Rattus rattus*. Contaminants of food are global problem, they cause economic damage and have a negative impact on food and feed i.e. human and animal's health.

Keywords: *mycotoxins, insects, health risks.*

Safe food means good health

Imperative of agriculture and processing industry is the production of safe food of high nutritional quality without the biological, physical and chemical contaminants. For secure safe final product is very important production and quality of raw materials i.e. "from farm to fork". Cereals and cereal products are very present in the nutrition of humans and animals (Perši *et al*., 2011). It is necessary to minimize the presence of contaminants in grain weight. Permanent companions of grain and grain mass are microorganisms. Contamination by microorganisms and their metabolites occurs in the fields, during the harvest, then during transport and storage (Coffey *et al.,* 2009). They pose a major problem especially expressed during the rainy weather (Pepeljnjak *et al.,* 2008; Mitak *et al*., 2011).

Fungi and mycotoxins as a serious danger

Fungi are ubiquitous plant pathogens that are major spoilage agents of foods and feedstuffs. The infection of plants by various fungi in the same time having results in reduction in crop yield and quality with significant economic losses. Also the fungi contribute contamination of grains with poisonous fungal secondary metabolites called mycotoxins. The ingestion of such mycotoxin contaminated grains by animals and humans has enormous public health significance. These toxins can cause diseases in human and animals (Makun, 2013). The disease produced by mycotoxins is termed as "mycotoxicosis" (Pal, 2007). Acute mycotoxicoses can cause serious and sometimes fatal diseases. The possibility of mycotoxin intoxication should be considered when an acute disease occurs in several persons when there is no evidence of infection with a known etiological agent, and no improvement in the clinical picture following treatment. Most of the outbreaks of mycotoxicoses described are a consequence of the ingestion of food that is contaminated with mycotoxins. The thorough control of food quality, in both industrialized and developing countries, is therefore necessary to avoid such outbreaks (Pal, 2015). According to Ožegović and Pepeljnjak (1995) - the most common molds are of genus *Aspergillus*, *Penicillium*, *Fusarium*, *Claviceps* and *Alternaria* and they are classified into field molds (*Alternaria*, *Helminthosporium*, *Fusarium* and *Cladosporium*) and storage molds (*Penicillium* and *Aspergillus*). Species of the genus *Penicillium*, *Fusarium* and *Alternaria*, as well as teleomorph class Ascomycetes (*Aspergillus alliaceus*, *Aspergillus nidulans*, etc.) are most often cited as potential producers of mycotoxins (Samson *et al*., 2004). *Aspergillus*, *Penicillium* and the *Eurotium* are "storage" fungi that develop within water activity (aw value) of 0.85 or lower, so that they can be isolated from spices, dried fruits, vegetables, sunflower seeds and similar products. Types from genus *Fusarium* and *Alternaria* are the "field" fungi and their development requires a higher moisture content in the substrate and lower temperature. These types are often found in/on grain cereals and cereal products (Kocić-Tanackov and Dimić, 2013). Mycotoxins are toxic byproducts (secondary metabolites) produced by fungi. Namely, aflatoxins, ochratoxin A, fumonisins, certain trichothecenes, and zearalenone are considered the most serious threat to human and animal health due to their carcinogenic potential of hepatogenic, teratogenic, and mutagenic effects (Pal, 2015).

Alternaria toxins biosynthesize different species of genus *Alternaria*, the frequent causal agents of plant diseases. These types of fungi are the main contaminants of wheat, sorghum, barley, sunflower, canola, tomatoes, apples, citrus fruits, olives (Desphande, 2002).

Zearalenone - zearalenone are like fumonisine derivatives polyketides. They are natural contaminants of harvested and stored grain and their derived products around the world (Kocić-Tanackov, 2004; Zinedine *et al.*, 2007; Weidenbörner, 2008; Rhyn and Zoller, 2003).

Sterigmatocystine (STC) is a secondary metabolite of some species of the genus *Aspergillus* and as the most important producer of this toxic metabolite is cited *A. versicolor* (Veršilovskis and De Saeger, 2010).

Aflatoxins are highly toxic coumarin derivatives mainly biosynthesized by *A. flavus* and *A. parasiticus*. The most important mycotoxins from this group are aflatoxins B1 (AB1), B2 (AB2), G1 (AG1), G2 (AG2), M1 (AM1) and M2 (AM2) (Samson *et al*., 2004; Park *et al*., 2000). These mycotoxins fungi biosynthesize in/on a number of substrates, such as oil seeds, cereals and their products, stone fruits, subtropical fruits, spices (Weidenbörner, 2008). Most often they are found in products that are not sufficiently dried after harvest or during storage at relatively high temperatures. From this group of mycotoxins AB1 is the strongest carcinogen, followed AG1, AM1 and AB2. At mammals aflatoxins cause acute aflatoxicosis, which are manifested primarily

through liver damage, although the kidneys, lungs and spleen may be damaged. AB1 is described as the strongest potential hepatocancerous (Fink-Gremmels, 2008).
The high average temperatures and a long drought cause heat stress in plants, causing a plant contamination with fungi of the genus *Aspergillus*, especially in the period of flowering and tanning corn silk (Marsh *et al.*, 1984; Kocić-Tanackov *et al.*, 2013).
Fusarium growth often implies production of mycotoxins (e.g. trichothecenes, zearalenone, fumonisins, moniliformin, etc) which pose a threat to human or animal health (Krsmanović *et* al., 2005).
One of the first known mycotoxicosis is ergotism. Ergotism was responsible for the deaths of thousands in medieval Europe (Duraković *et al*., 2003; Diaz, 2005).
Ergot alkaloids are compounds produced as a toxic mixture of alkaloids in the sclerotia of species of *Claviceps*, which are common pathogens of various grass species. Modern methods of grain cleaning have significantly reduced ergotism as a human disease. However, it is still an important veterinary problem (Bennett and Klich, 2003).
The most important in the transmission of mycotoxins are grains because they consume humans and animals. It is assumed that even 25 to 40% of the world grains are contaminated with mycotoxins (Pittet, 1998; Peraica and Domijan, 2001, Surai *et al*., 2008). Although there are more than 300 isolated mycotoxins (Betina, 1984), studies mainly refer to those who threaten the health of people, domestic animals and pets. In recent years numerous studies have been conducted on the impact of mycotoxins on human health and the emergence of certain diseases of unclear etiology. Since 1993, "World Health Organization International Agency for Research on Cancer" estimated potential carcinogenic and mutagenic effects of certain mycotoxins on humans (WHO-IARC, and WHOIARC 1993a, 1993b). Comprehensive negative effect on people (Peraica *et a*l., 1999) and animals (D'Mello and MacDonald, 1997) gave in their review articles.
Crop rotation (Jaime Garcia and Cotty, 2010) is one way to reduce the risk of contamination by molds and mycotoxins in the field. Alsao, the storage conditions after harvest are of great importance for the prevention of occurrence of mycotoxins. Sorting and drying kernels are important parameters to prevent subsequent contamination of grain mycotoxins (Jaime Garcia and Cotty, 2007; Birck *et al*., 2006).

Insects as vectors of disease

Stored products of agricultural and animal origin are attacked by more than 600 species of beetle pests, 70 species of moths and about 355 species of mites causing quantitative and qualitative losses (Rajendran, 2002). Insect contamination in food commodities is an important quality control problem of concern for food industries. In industrialized countries like Canada and Australia there is zero tolerance for insects in food grains (White, 1995; Pheloung and Macbeth, 2002). The fact is that a reduction in quality and contamination of food by insects, their metabolites and excrements makes food unsuitable for human consumption. The most common insects as food contaminants negatively affect the quality of the food itself, but also on human health (Orris and Whitehead, 2000). Insects cause physical, chemical and microbiological contamination of products in warehouses. Physical contamination represent occurrence of different parts of the insect body or insect stage (eggs, larvae, exuviae or excrements). Many insect species in warehouses are hosts and vectors of pathogens of man and/or animals. In that way become leading agents in microbial contamination of stored food products (Olsen, 1998). The most common contaminants are the insects with hidden development - within the stored grains such as *Rhyzopertha sp*. and *Sitophilus sp* (Tanasković *et al*). *Sitophilus oryzae* (L.), *S. granaries* (L.) or *Rhyzopertha dominica* (F.) are the main source of physical contamination in

flour (Pedersen, 1992; Perez-Mendoza *et al*., 2003). The major storage pests of milled rice are *Sitophilus spp*. and *Tribolium caslaneum*, with *Corcyra cephalonica*, *Ephestia kuehniella*. *Rhyzopertha dominica* and some other secondary pests causing minor damage (Sidik *et al*., 1985). Carcinogenic and teratogenic substances are produced only by *Tribolium spp*. (El-Mofty *et al*., 1989, 1992). Namely, in secretion of these species (*T. castaneum*) were identified 13 different quinone (Howard, 1987). These ingredients give an unpleasant flavor in storage products and at some small vertebrates can cause cancer of the liver and spleen (El-Mofty *et al*., 1988; 1989; 1992). Although quinones may endanger the health of animals for experiments, the level of contamination *T. castaneum* in warehouses of finished products is low or normal, however cumulation quinone may endanger human health (Hodges *et al*., 1996). Insects of ranks Blattodea, Coleoptera, Lepidoptera and Psocoptera can cause allergenic reactions in humans. Sources of allergies are body parts or secretions of insects and mites. Continuous exposure to insects and mites can cause occupational diseases in farmers, millers and bakers (Revsbech and Dueholm, 1990). Exposure of infested grain dust is associated with a number of allergic reactions to a type of conjunctivitis, rhinitis, dermatitis, and asthma (Jeebhay *et al*., 2001).

Rodents attacks for humans and animals health

Since the Middle Ages is known that rodents can contribute to human disease, as black rats were associated with distribution of plague. However, also in modern times rodents form a threat for public health (Meerburg *et al*, 2009). Rodent-related problems exist in public health due to their role as vectors and reservoirs for bacterial diseases such as leptospirosis, murine typhus and salmonellosis, in addition to plague. As humans and livestock are in regular contact with rodents, the potential for transmission of zoonotic diseases is high (Gratz, 1994; Webster and Macdonald, 1995). Because of their adaptable characteristics, types of commensal rodents, house mouse (*Mus musculus*) and the brown rat (*Rattus norvegicus*) have adapted to the conditions of storage areas, which are easily accessible shelters and food (Vukša, 2010). Except with their nutrition, rodents contaminate up to nine times more food than they need for food with their feces, urine, hair, and many other impurities (Drummond, 2001; Brown et al., 2007).

Detection of contaminants

Mycotoxins in feeds, milk, urine, blood, bile, and faeces can be detected by several techniques such as TLC, GLC, HPCL, ELISA, RIA, and RFLP (Pal, 2015).

To identify a physical contamination by insects may be used flotation fragments, immunochemical methods (ELISA), molecular methods (PCR) and infrared spectroscopy - NIRS (Perez-Mendoza *et al*., 2005).

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