



MICROBIAL CONTAMINATION OF THE AIR IN LIVESTOCK BUILDINGS AS A THREAT TO HUMAN AND ANIMAL HEALTH – A REVIEW

Anna Chmielowiec-Korzeniowska¹, Beata Trawińska¹, Leszek Tymczyna^{1*}, Hanna Bis-Wencel¹,
Łukasz Matuszewski²

¹Department of Animal Hygiene and Environmental Hazards, University of Life Sciences,
Akademicka 13, 20-950 Lublin, Poland

²Medical University in Lublin, Gębali 6, 20-093 Lublin, Poland

*Corresponding author: leszek.tymczyna@up.lublin.pl

Abstract

Livestock buildings are often contaminated with bacterial and fungal microflora. Animals living in the buildings, especially their excreta and secretions and their feed, can be a source of microorganisms, including pathogens. Significant microbial contamination occurs in pig houses, poultry houses and cowsheds. The microbes most frequently isolated from the air of these buildings are bacteria of the genera *Streptococcus*, *Staphylococcus*, *Bacillus*, and *Clostridium* and of the family *Enterobacteriaceae*. Among fungi, the most common are *Aspergillus*, *Trichoderma*, *Penicillium*, *Cladosporium* and *Alternaria*. Microbes present in livestock buildings often pose a hazard to workers, in whom they can cause infectious and allergic diseases, especially respiratory disease. Bacterial endotoxins may also pose a threat to humans and animals. For this reason it is important to carry out microbiological monitoring and preventive measures on livestock farms and to maintain appropriate environmental conditions. This will reduce microbiological contamination of livestock buildings and improve both workers' health and animal welfare.

Key words: livestock housing, bacteria, fungi, endotoxins, human and animal health hazard

The composition of the air of livestock buildings differs significantly from that of atmospheric air. The air in buildings housing animals is contaminated with decomposition products of organic matter – alcohols, aldehydes, amines and organochlorides, as well as compounds formed during the metabolism of sulphur-containing amino acids in the animals' digestive system, i.e. sulphides and mercaptans (Chmielowiec-Korzeniowska et al., 2018). Microbial contaminants are a permanent component of the livestock farming environment, due in part to the high humidity in the buildings and higher air temperature than that of the air outside. These contaminants are present in the air in the form of biological aerosols and can play an important role in the

transmission of allergic and infectious diseases, even contributing to epizootics. The qualitative and quantitative composition of air undergoes significant changes in time and space. Regarding the type and size of bioaerosol particles forming the dispersed phase, the air of livestock buildings most often contains viruses ($<0.03\ \mu\text{m}$), bacteria ($0.25\text{--}8\ \mu\text{m}$), fungal spores ($1\text{--}33\ \mu\text{m}$) and pollen ($17\text{--}58\ \mu\text{m}$) (Dungan and Leytem, 2009), as well as toxins (endotoxins, peptidoglycans and mycotoxins) and fragments of plant and animal tissues (Yang et al., 2018).

In the case of bacteria suspended in the air, as many as 80% occur in the form of conglomerates, i.e. microbes that adhere to dust particles (Pejsak, 2007), so their number increases with the dust concentration in the air (Nimmermark et al., 2009). Mesophilic bacteria predominate among microorganisms in the air of livestock buildings (Dutkiewicz and Górny, 2002). Most of these are saprophytic bacteria, although some may be pathogenic or conditionally pathogenic. Bacteria in bioaerosols can not only cause infectious diseases, but also have immunotoxic and allergic effects.

It is primarily Gram-positive bacteria that are associated with bioaerosol particles. In the total pool of identified bacteria, Gram-negative bacteria constitute only 10% (pig houses), 4.9% (cowsheds) and 2.6% (henhouses) (Bakutis et al., 2004). Their number increases in buildings contaminated with organic dust. Bioaerosols also contain fungi, especially mould fungi, which are a source of toxic mycotoxins. Poisoning with these metabolites is a serious nutritional and veterinary problem (Tymczynyna and Bartecki, 2007).

Endotoxins, high-molecular-weight lipopolysaccharides (LPS) present in the outer layer of the cell wall of Gram-negative bacteria, pose a particular threat. These substances, which exhibit high biological activity, can be a potential cause of respiratory disease in people inhaling them. Endotoxins are among the most ubiquitous organic compounds in agriculture. This is partly due to their heat resistance, which allows them to survive in the environment even in extreme conditions (Wassenaar and Zimmermann, 2018). Disinfection reduces the number of microorganisms, but does not remove their residues. Both dividing and dead Gram-negative bacteria release LPS.

Sources of microbial air contamination

Animals, especially their excreta and secretions, are the main source of microbial air contamination (Chmielowiec-Korzeniowska et al., 2018; Ropek and Frączek, 2016; Skóra et al., 2016; Szadkowska-Stańczyk et al., 2010). Molecular biology methods used by Nehme et al. (2008) confirm that pig faeces are the main source of microbial air contamination in pig houses. Excreta under animals are a rich reservoir of Gram-positive cocci of the genera *Streptococcus*, *Peptostreptococcus*, and *Staphylococcus*, bacteria of the genera *Lactobacillus* and *Bacillus*, and Gram-negative *Escherichia* bacteria (Zhu, 2000). Enterohaemorrhagic *E. coli* O157:H7 serotypes, which cause haemorrhagic colitis in humans, may also be found in the solid or liquid manure of cattle, and less often of pigs and poultry (Lim et al., 2010; Oporto et al., 2008). Pathogenic microorganisms such as *Salmonella* spp., *Campylobacter* spp., *Listeria monocytogenes*, or *Yersinia enterocolitica* from diseased animals or carriers can also be found here (Manyi-Loh et al., 2016; Myszograj and Puchalska, 2012).

Many authors emphasize the relatively long survival time of these microorganisms in solid and liquid manure, which poses a serious risk of transmission to the natural environment (Cotta et al., 2003; Hutchison et al., 2005; Kostadinova et al., 2014; Lu et al., 2003; Manyi-Loh et al., 2016; Zhu, 2000)

Litter is another important source of biological air contamination. Lu et al. (2003) examined the microbial composition of poultry litter and showed a very high average bacterial concentration of 10^9 CFU/g. Intestinal bacteria, including *Enterococcus* spp. and coliforms, constituted only 0.1% and 0.01% of the total number of microorganisms, while the dominant species were *Globicatella sulfidofaciens*, *Corynebacterium ammoniagenes*, *Corynebacterium urealyticum*, *Clostridium aminovalericum*, *Denitrobacter permanentens*, and bacteria of the genus *Arthrobacter*. Pathogenic bacteria that were identified in manure were not detected in the litter, e.g. *E. coli*, *Salmonella* spp. and *Campylobacter* spp., which the authors believe could be due to the less favourable microenvironment of faeces mixed with litter compared to faeces alone.

Microorganisms also enter the air from feed and microbiologically contaminated water. Research confirms that feed and drinking water contaminated with faeces of sick animals or carriers can be an important source of pathogenic strains of *Salmonella* spp. and *E. coli* (Cabral, 2010; Truszczyński and Pejsak, 2015). High temperature and high humidity are conducive to their multiplication (Budzińska et al., 2014; Yao et al., 2010).

Quantitative and qualitative composition of bioaerosols in livestock buildings

Quantitative composition

Air microflora has been the subject of numerous scientific studies in recent years. Research work has primarily aimed at quantitative and qualitative characterization of microorganisms found in the air, as well as identification of the factors affecting its quality. In most studies, the total bacterial concentration exceeded recommendations regarding microclimate conditions for livestock (8.0×10^4 CFU/m³) (Kończak and Dobrzański, 2006) as well as hygiene recommendations, determined according to the criterion of the health of people working in this sector (1.0×10^5 CFU/m³ for mesophilic bacteria, 2.0×10^4 CFU/m³ for Gram-negative rods, 5.0×10^4 CFU/m³ for fungi, and 2000 EU/m³ for endotoxins (Górny et al., 2011).

Henhouses have the highest levels of bacterial and fungal bioaerosols compared to livestock buildings housing other animals. This is confirmed by Bakutis et al. (2004) and Stuper-Szablewska et al. (2018). An important factor increasing the concentration of microorganisms is the dust concentration, which is much higher in poultry houses than in other buildings. Skóra et al. (2016), in an assessment of the content of microorganisms in settled dust collected from henhouses, showed that the concentration of bacteria was as high as 3.2×10^9 CFU/g, while that of fungi was 1.2×10^6 CFU/g. Such a high level of microbial contamination also contributes to a very high level of endotoxins (Schierl et al., 2007; Stuper-Szablewska et al., 2018). In the case of pig houses, the LPS concentration usually does not exceed a few hundred thousand endotoxin units per m³ of air (Chmielowiec-Korzeniowska et al.,

2018; Szadkowska-Stańczyk et al., 2010). Contamination of the air of buildings for cattle is lower, and its level is determined by the production type – meat or dairy (Bakutis et al., 2004). Abd-Elall (2009) showed that the average number of bacteria in the air of buildings for beef cattle was higher than in cowsheds housing dairy cattle. Matković et al. (2006) also found variations in bacterial aerosol concentrations over the course of a day. The highest values were noted in the evening, with a maximum level of 7.76×10^4 CFU/m³.

Bioaerosol species composition

The composition of air microflora depends on the species of livestock and above all on the animals' health, microclimatic and hygiene conditions, and the quality of bedding and feed (Budzińska et al., 2014; Chang et al., 2001).

Gram-positive bacteria, i.e. bacteria of the genera *Staphylococcus*, *Corynebacterium* and *Streptococcus*, which constitute the physiological microbiota of the skin and mucous membranes of animals, predominate in the air of livestock housing (Seedorf et al., 1998). In pig houses, Gram-positive bacteria are mainly represented by the genus *Staphylococcus*, including *S. aureus*, *S. cohnii*, *S. urealyticus* and *S. intermedius* (Roque et al., 2016), as well as the genera *Aerococcus*, *Micrococcus* and *Bacillus* (Predicala et al., 2002). Among Gram-negative bacteria, the most common are *Escherichia coli*, *Enterobacter agglomerans*, *Acinetobacter baumannii* (Zucker and Müller, 2000), and bacteria of the genera *Moraxella* and *Pseudomonas* (Predicala et al., 2002). In addition to these bacteria, pathogenic and conditionally pathogenic bacteria may be present in the air of the pig house, including the aforementioned *Staphylococcus aureus* and *Streptococcus suis*, *Pasteurella* spp., *Bordetella* spp., *Actinobacillus* spp., *Salmonella* spp. and *Mycoplasma* spp. (Zhu, 2000).

Bacteria of the genera *Staphylococcus*, *Streptococcus* and *Bacillus* are also the most common in henhouses, as well as bacteria of the genus *Clostridium* (Hartung and Schulz, 2011; Stuper-Szablewska et al., 2018).

Roque et al. (2016), determining the microbial composition of the air of cowsheds, usually identified Gram-positive bacteria, including *Bacillus cereus*, *B. licheniformis*, *Enterococcus faecalis*, *Staphylococcus chromogenes* and *S. lentus*. The authors isolated only one species of Gram-negative bacteria: *Acinetobacter lwoffii*.

Mycological air contamination consists of the allergenic and toxin-producing fungi *Aspergillus* spp., *Penicillium* spp., *Cryptococcus* spp., *Cladosporium* spp. and *Alternaria* spp., as well as yeasts of the genus *Candida* (Stuper-Szablewska et al., 2018).

The widespread use of antibiotics in animal farming has led to the increasing occurrence of drug-resistant microorganisms in this environment. Strains resulting from mutation pressure exerted by an antibiotic or gene transfer from bacteria that have previously developed resistance mechanisms constitute a significant percentage of isolated bacteria (Szewczyk et al., 2019). The main problem is strains of Gram-negative bacilli, especially from the family *Enterobacteriaceae*, producing extended-spectrum β -lactamases (ESBL), AmpC cephalosporinases, and carbapenemases. These strains show resistance not only to β -lactam drugs (Nikonorow et al., 2013). This phenomenon is most commonly noted in enteropathogenic bacteria, primarily *Salmonella* spp., *E. coli*, and *Campylobacter* spp.

The high frequency of emergence of strains resistant to two or more antibiotics (multi-drug resistance) is worrying. The presence of multi-drug resistant bacterial strains in pig faeces, including *Salmonella* (58% of isolated strains), *E. coli* (13.4%), and Gram-positive *Enterococcus faecalis* and *Enterococcus faecium* (19.6% and 25.3%), has been reported by Wegener et al. (2003) and Hölzel et al. (2010).

Bacteria resistant to several antibiotics have also been isolated from samples collected from cattle and poultry (Lee, 2006; McEachran et al., 2015), as well as from food products derived from them, i.e. meat (Kitai et al., 2005; Normanno et al., 2007), milk, and dairy products (Peles et al., 2007; Keyvan et al., 2020). Wasyl et al. (2013) emphasize that the level of intake of antimicrobials has a marked effect on the number of drug-resistant bacteria isolated, and therefore resistance is most widespread in broiler and turkey housing and less prevalent in cowsheds.

Non-therapeutic use of antibiotics in animal production is another important factor responsible for the emergence of methicillin-resistant *S. aureus* strains, known as LA MRSA (livestock-associated methicillin-resistant *S. aureus*), isolated mainly from pigs and calves (Normanno et al., 2007). Friesea et al. (2012) have also demonstrated the frequent presence of LA MRSA in dust contaminating the air of livestock buildings, in which they can survive for many months.

Factors affecting the quantitative and qualitative composition of bioaerosols

Microbial contamination of the air of livestock buildings largely depends on the stocking density, type of diet, housing, and environmental conditions (Budzińska et al., 2014; Chang et al., 2001; Zhao et al., 2014).

Animals

The number of microorganisms present in the air is closely linked to the stocking density, age and weight of animals, and their activity. Concentrations of microorganisms and dust in the air generally increase with the age and body weight of animals. Emissions of gaseous contaminants increase as well (Cambra-López et al., 2010; Nimmermark et al., 2009; Stuper-Szablewska et al., 2018). As the amount of excrement increases, the number of microorganisms released increases with it (Linlin et al., 2018), while prolongation of the fattening period reduces moisture in the litter. This increases the amount of dust in the air, and thus affects the number of microorganisms in it (Cambra-López et al., 2010).

The concentration of bacterial and fungal aerosol, as well as endotoxins and β -glucans, increases with each stage of production. This is linked to the animals' growth as well as to microclimate conditions specific to a given cycle. A change in temperature inside the building entails a change in the bioaerosol composition and the rate of growth and multiplication of microorganisms (Lawniczek-Walczyk et al., 2013; Oppliger et al., 2008).

Research by Yang et al. (2018) has shown a direct cause-and-effect relationship between animal activity and dust concentration. A wider scope of animal activity increases bioaerosol formation, as confirmed by Saleh et al. (2005). The decrease in the concentration of microorganisms and dust in the late period of broiler fattening can be linked to the reduced activity of larger, heavier birds. Cambra-López et al. (2010), in a study conducted in a pig house, showed that the activity of pigs, and thus the

amount of microbial particles released into the air, decreased as they grew. In poultry, due to the higher activity of laying hens during the day, the dust concentration seems to be more linked to changes over the course of the day, and in broiler houses with the lighting programme (Cambra-López et al., 2010).

Microbial contamination of livestock buildings, especially its qualitative composition, is determined by the animals' state of health. Sick animals or carriers can be a source of pathogenic air microflora (Pejsak, 2007).

Microclimatic conditions

Microbial air contamination in livestock buildings is highly dependent on temperature and humidity, ventilation efficiency, and the quality and physicochemical properties of litter. The type of ventilation, age of buildings, and type of litter also have a significant impact on air contamination in poultry houses (Banhazi et al., 2008).

The microclimatic conditions of buildings, including relative humidity and dust concentration, are closely correlated with the fungal composition of the air (Kumari et al., 2016). A positive correlation has also been found between a high temperature in the pig house and the total number of coliforms and *E. coli* (Yao et al., 2010).

High temperature and humidity promote the multiplication of microorganisms in buildings where animals are kept (Lonc and Plewa, 2010).

High humidity has a positive effect on the survival and/or reproduction of species of microorganisms sensitive to dehydration. On the other hand, in a humid environment, dust particles bind to the surface more easily, which impedes their rise and causes them to settle faster (Zhao et al., 2014). Cambra-López et al. (2010) report that relative humidity above just 70% facilitates aggregation of particles suspended in the air, causing them to settle faster, which reduces the dust concentration and biological air contamination.

Although the litter system of livestock housing improves animal welfare, it entails higher emissions of microbial contaminants and may cause more health problems in the animals. Increased air movement above the litter, which occurs during the animals' physical activity and the operation of fans, increases the amount of contaminants released (Zhao et al., 2014). The type of bedding material used and its hygienic condition are not insignificant (Banhazi et al., 2008). The number of bacteria in the air of poultry houses often increases, especially in the case of birds kept for a prolonged period of time, because the fermentation rate in the manure increases, and alkalization of the litter promotes the growth of microorganisms. On the other hand, Kostadinova et al. (2014) demonstrate that fresh manure usually has a higher number of microbes and of coliforms than litter, and their number decreases together with the moisture level in the excreta under the animals.

Season

The bioaerosol concentration in farm buildings is influenced by the season of the year, which is inextricably linked to ventilation parameters (Budzińska et al., 2014; Nehme et al., 2008; Popescu et al., 2014). When temperatures are above freezing, increased ventilation and the resulting greater air turbulence cause an increase in bioaerosols (Cambra-López et al., 2010). Wójcik et al. (2010), assessing microbial air contamination in barns for dairy cows kept in a free-stall system, found the highest

number of mesophilic aerobic bacteria and fungi in spring, compared to the summer, autumn and winter. Ropek and Frączek (2016) observed the reverse relationship. They noted the highest microbial contamination in the air of cowsheds in autumn and winter, because in summer and spring the animals grazed on pastures and spent less time indoors.

Chmielowiec-Korzeniowska et al. (2018) and Budzińska et al. (2014), showed no dependence of the bioaerosol concentration in a pig house on the time of year. The number of bacteria and fungi was similar irrespective of the outside temperature, and differences were found only in their species composition.

Health risk for people and animals

Microbial contaminants accumulated in closed livestock buildings are a serious health risk for the animals and workers (Stuper-Szablewska et al., 2018; Szadkowska-Stańczyk, 2010), and when released outside by the ventilation system, they can pose a threat to the natural environment and the health of people living in the area (Chmielowiec-Korzeniowska et al., 2018; Plewa-Tutaj et al., 2014). These hazards are caused not only by microorganisms suspended in the air (bacteria and fungi) but also by the residues they produce (toxins).

Bacteria

Allergens, immunotoxins and infectious agents, including pathogenic microorganisms inducing zoonoses and drug-resistant strains, pose the greatest threat to people handling livestock (Dutkiewicz and Górny, 2002). In livestock farm workers, these factors most often cause respiratory diseases, including chronic bronchitis and obstructive pulmonary disease, as well as asthma and organic dust toxic syndrome (ODTS) (Hartung and Schulz, 2011; Hamid et al., 2018; May et al., 2012; Viegas et al., 2013). Research results have confirmed that every fifth worker employed in agriculture suffers from respiratory disorders, with symptoms such as wheezing and shortness of breath (Hamid et al., 2018).

The main threats to the health of industrial poultry farm workers are *Chlamydia* spp. (causing interstitial pneumonia), *Listeria monocytogenes* (causing listeriosis in humans and animals), *Staphylococcus aureus* (purulent infections, airway inflammation, and even sepsis in people with low immunity), *Streptococcus* spp. (infections of various organs, including the urinary tract, lungs, and mouth), *Salmonella* Enteritidis, and *S. Typhimurium* (causing salmonellosis in humans and animals) (Myszograj and Puchalska, 2012). The air in cowsheds, pig houses and stables may also contain rods from the family *Corynebacteriaceae*, causing allergic diseases in humans, including allergic alveolitis (Dungan, 2010). In addition, bacteria from the family *Enterobacteriaceae* have a harmful effect on workers' health, because long-term exposure to their endotoxins can also contribute to the onset of allergic alveolitis and asthma (Oppliger et al., 2008). Research carried out in cowsheds has shown that microorganisms present in the air of housing for meat cattle pose a greater danger to humans than those in housing for dairy cattle (Abd-Elall, 2009).

Air contamination in livestock buildings also has a detrimental effect on the animals kept in them. The most common respiratory diseases result from a combination of many factors, including chemicals and/or microorganisms present in the air. In

the air of pig houses, an excellent example is porcine respiratory disease complex (PRDC), whose aetiological factors are various types of bacteria and/or viruses and unsuitable environmental conditions to which the pig's respiratory system is exposed throughout the fattening period (Loera-Muro, 2015).

A study covering 28 pig farms was carried out in Sweden to assess the correlation between air contamination in pig housing and diseases and productivity in pigs. The most commonly identified health problems in these animals were pneumonia, pleurisy, and increased neonatal mortality (Donham, 1991).

The combined effect of air contaminants is considered to be one of the most important stressors in the living environment of animals. High concentrations of microorganisms, endotoxins, mycotoxins, gas admixtures and dust adversely affect the structure and defence functions of mucous membranes, especially the respiratory mucosa and the conjunctiva, causing allergic reactions and inflammation and increasing the incidence of infectious diseases in animals (Plewa-Tutaj et al., 2014). In addition, the emergence of antimicrobial resistance in commensal bacteria or animal pathogens contributes to an increase in the numbers of resistance genes that can be transmitted to human pathogens. Despite bans on the non-therapeutic use of antibiotics in animals, the effectiveness of prevention and treatment of an increasing number of infections continues to decrease. The scale of the problem can be seen in a report by the World Health Organization on the absolute necessity of reducing the spread of antibiotic resistance from agricultural sources (WHO, 2014).

Fungi

The negative impact of moulds of the genera *Aspergillus*, *Penicillium*, *Fusarium*, *Stachybotrys* and *Trichoderma* on the health of humans and animals is well known. Fungi found in the air of livestock rooms, including those of the genera *Cladosporium* and *Alternaria*, can cause allergic reactions, asthma and conjunctivitis, and may also contribute to the occurrence of mycoses, especially in the organs. Due to their small size, fungal spores can very easily reach the lower respiratory tract. Epidemiological studies have shown that the most common causes of airborne allergies are fungi of the genera *Alternaria* and *Cladosporium*, as well as *Penicillium* and *Aspergillus*. Fungi of the genera *Mucor*, *Botrytis*, *Fusarium*, *Pullularia*, *Curvularia*, *Phoma* and *Rhizopus* are less likely to cause allergic reactions (Żukiewicz-Sobczak et al., 2013).

Depending on the degree of invasiveness and individual resistance, these fungi can also cause a wide range of pulmonary diseases. Fungi of the genus *Penicillium* commonly found in the natural environment but also in raw material of plant origin, are recognized as a factor in allergic rhinitis and allergic bronchopulmonary mycoses (Dutkiewicz et al., 2011). Inhalation of large quantities of fungal antigens in organic dust can also lead to the development of allergic alveolitis. This condition may be caused by inhalation of organic dust originating in mouldy hay or feed contaminated with Actinobacteria and fungi of the genus *Aspergillus* (Sabino et al., 2019).

The genus *Aspergillus* includes species that can induce allergic reactions of the respiratory tract (*A. candidus*, *A. niger* and *A. versicolor*), allergic alveolitis (*A. clavatus*, *A. flavus*, *A. fumigatus* and *A. terreus*), allergic rhinitis (*A. fumigatus*), bronchial asthma (*A. flavus*, *A. fumigatus* and *A. oryzae*), and respiratory infections,

such as pulmonary mycosis (*A. flavus*, *A. fumigatus* and *A. terreus*). Immunotoxic effects (*A. candida*) and toxic effects induced by secreted mycotoxins (*A. fumigatus*, *A. niger*, *A. ochraceus*, *A. oryzae*, *A. terreus*, and *A. versicolor*) have been demonstrated, as well as carcinogenic effects (*A. flavus*).

Toxins

Air contaminated with microorganisms, and especially their residues, can cause enzyme imbalance in the body. Inhaled bioaerosols together with the substances they produce (endotoxins, peptidoglycans, glucans and mycotoxins) exert an immunotoxic effect on the immune system of the lungs (Dutkiewicz et al., 2011).

LPS toxicity is manifested by fever, a change in leukocyte count, difficulty breathing, and even death. Endotoxins inhaled by a human being together with dust non-specifically activate pulmonary macrophages, leading to overproduction of mediators, a drop in blood pressure, high fever, disseminated intravascular coagulation, and lethal septic shock (Tymczyzna and Bartecki, 2007).

Research carried out in Switzerland showed that people working in poultry houses, especially at the end of the fattening period, were significantly exposed to bioaerosols containing endotoxins. During this period the concentration of endotoxins in the air exceeded 6,000 EU/m³. This value was six times higher than recommended, increasing the likelihood of respiratory disorders (Oppliger et al., 2008).

Similar observations apply to the animals themselves. Linlin et al. (2018) showed that pigs kept in buildings with high levels of endotoxins had higher total leukocyte, neutrophil and lymphocyte counts in the peripheral blood than those with low endotoxin exposure. Kullik et al. (2013) found that intravenous injection of pigs with lipopolysaccharide (LPS) significantly increased plasma TNF α and IL-6 levels. Tang et al. (2019) emphasize that high concentrations of endotoxins in inhaled air can induce cell death, production of reactive oxygen species (ROS), and the release of inflammatory cytokines (IL-1 β , IL-18, TNF- α) and cyclooxygenase-2 (COX-2) by activating the TLR4/MyD88 and NLRP3 inflammasome pathways. Roque et al. (2018) studied pigs exposed to high doses of endotoxins and found an increase in the number of circulating neutrophils and lymphocytes and in levels of IgG and IgE immunoglobulins and interferon-gamma (IFN γ), and a decrease in the level of immunoglobulin A and tumour necrosis factor TNF α . Continuous exposure of pigs to high levels of endotoxins in the air can lead to abnormal immune profiles. Moreover, a study by Skórska (1991) showed that the presence of endotoxins in inhaled air may impair the reproductive system of experimental animals and cause changes in sexual behaviour. High exposure causes morphological changes in the Leydig and Sertoli cells, as well as impairment of spermatogenesis. It has been observed to increase the number of primary and secondary sperm defects, decrease sperm concentration and motility, and cause hormonal changes (testosterone and 17 β -estradiol) in the blood and sperm of exposed animals.

Mycotoxins produced by fungi, of which the best known are aflatoxins, ochratoxins, trichothecenes and zearalenone, cause dermatitis; acute intoxication with headache; diarrhoea; immune mechanism impairment; and liver and kidney damage in humans. Mycotoxins also interfere with the functioning of the nervous system. Many

of them, including aflatoxin, are teratogenic, mutagenic and carcinogenic (Stuper-Szablewska et al., 2018).

Mycotoxins also adversely affect animal productivity and health (Akinmusire et al., 2019; Kanora and Maes, 2009; Murugesan et al., 2015). They can be harmful to various organs in poultry, primarily the gastrointestinal tract, liver and immune system, even leading to death in extreme cases (Murugesan et al., 2015). Research by Kanora and Maes (2009) demonstrates that toxins produced by *Aspergillus* spp., *Fusarium* spp. and *Claviceps* spp. can also impair fertility and reproduction in sows.

Moulds are a source not only of mycotoxins, but also of volatile organic compounds (e.g. aldehydes and ketones) that cause intoxication with systemic symptoms, as well as immunotoxic β -1,3-glucans, which are an important factor in the development of lung disease, both inflammatory and allergic (Dutkiewicz et al., 2011).

All of these factors acting on the animal body for a long time disturb local homeostasis. The relationship between excessive stimulation of the immune system and inhibition of animal growth is well known (Kończak and Bodak, 2000). Cytokines released during the acute phase integrate the immune response at the neuroendocrine level with modulation of neurotransmitters and hormones (GH, IGF-1, CRH, ACTH, glucocorticoids, prostaglandins, and catecholamines), the main growth regulators. Behavioural symptoms of activation of these factors include reduced food intake and slowed digestion. In states of immune stimulation, catabolic processes are increased with a simultaneous decrease in tissue protein synthesis and increased generation of acute phase proteins, resulting in a negative energy and nitrogen balance. When there is a deficiency of metabolic amino acids needed for synthesis of immune proteins, uptake of structural amino acids increases. The loss of muscle proteins not only reduces production efficiency, but also adversely affects carcass value (Kończak and Bodak, 2000).

Conclusions

Modern animal production should not only provide economic benefits, but also take into account animal welfare and the natural environment. A condition for obtaining a high level of production is the health of the livestock. Any areas of neglect that reduce animal welfare can interfere with natural behavioural responses, inhibit growth and development, and above all increase the incidence of disease and death. Maintaining a low level of immune stimulation in animals by creating optimal living conditions with a low number of pathogens increases the amount of feed consumed, which results in a better growth rate and raises production efficiency.

Due to the potential threat to human and animal health on farms, it is essential to carry out microbiological monitoring and appropriate biosecurity measures in livestock housing. These include prophylaxis and maintenance of proper microclimatic conditions in livestock buildings. Attention should be paid to proper ventilation, temperature and relative humidity, and the buildings should be sanitized and disinfected. These measures reduce the level of microbial contamination of livestock buildings, and thus improve animal welfare and employee health. In addition, the introduction of restrictions on the amount of drugs used, accompanied by optimization of farming

methods, can reduce the number of antibiotic-resistant microorganisms posing such a great threat to public health.

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