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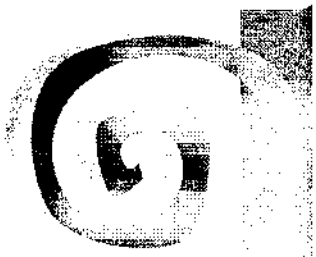

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INFLUENCE OF MYCOTOXINS IN SWINE FEED ON THE HEALTH STATUS OF PIGLETS

Prodanov-Radulović Z. Jasna*[1], Došen Đ. Radoslav[1], Stojanov M. Igor[1], Pušić M. Ivan[1], Živkov-Baloš M. Milica[1], Urošević I. Mirosłav[2]

[1]Scientific Veterinary Institute „Novi Sad", Rumenacki put 20, 21000 Novi Sad, Serbia;
[2]Agriculture School, Carice Milice 2, Futog, Serbia

*Corresponding author:
Phone: +381214895313
Fax: +38121518544
E-mail address: jasna.prodanov@gmail.com

ABSTRACT: Mycotoxins are secondary metabolites of fungi that can contaminate animal feeds at all stages of food production chain. Consumption of feed contaminated with mycotoxins may result in immunosuppresion, which represent a factor predisposing livestock to infectious diseases. From the epidemiological point of view, it is important to note that mycotoxins may cause breakdown of active immunity and occurence of disease even in properly vaccinated animals. The aim of the paper was to evaluate the influence of mycotoxins on the health status of piglets. The material for this research included the samples from five swine farms, where health disorders in suckling and weaned piglets were detected. Depending on the specificity of each evaluated case and available material, the applied research methods included: epidemiological and clinical evaluation, pathomorphological examination, standard laboratory testing for detection the precence of aerobic and anaerobic bacteria, virological testing and microbiological feed testing, in order to examine the presence of fungi and mycotoxins by the method of thin layer chromatography. In our research the persistant presence of various infections, which react poorly or do not react on the applied antimicrobial therapy was discovered. The presence of mycotoxin in feed can be directly connected to the detected health disturbances in piglets on the examined swine farms (vulvovaginitis, pneumonia, gastroenteritis)
Key words: *mycotoxins, swine feed, piglets diseases*

INTRODUCTION

Mycotoxins are secondary metabolites of mould, and so far, approximately 400 secondary metabolites with toxigenic potential produced by more than 100 moulds have been reported (Kabak et al., 2006). At the global level, it is considered that 25% of the world crop production is contaminated by mycotoxins, which may be a risk factor affecting human and animal health (Bouhet and Oswald, 2005). The issue of mycotoxins and mycotoxicoses in veterinary medicine is directly connected to the usage of mouldy and adversely storage of diffent types of grains (corn, wheat, barley) in animals feed (Osweiler, 2006).
In swine production in our geographic region, the most common are mycotoxicosis caused by zearalenon (F-2 toxin, ZEA), but also aflatoxins (AF), ochratoxin (OCT) and trichothecenes. (Gonzales and Rodriguez, 2008). The clinical and patomorphological picture of mycotoxicoses in swine depend on the age and category (breeding animals, suckling and weaned piglets, fatteners) (Prodanov et al., 2009). The young animals are much more sensitive to the effects of mycotoxins comparing to adults. Mycotoxins have hepatotoxic, nephrotoxic and immunosuppressive effects (Kabak et al., 2006), which can further complicate clinical and pathomorphological picture and diagnosis of mycotoxicosis in swine. The biggest challenge with mycotoxicoses is the non-specific nature of symptoms in the affected animals. Consequently, the health disorders due to mycotoxins in the feed are difficult to diagnose (Osweiler, 2006; Prodanov et al, 2009). It has been recognised by veterinary clinicians that marked immunosuppression is observed in livestock ingesting mycotoxins at levels below those that cause overt toxicity (Oswald et al., 2005). Immunosuppressive effects of mycotoxins are of special interest and may have significant influence on the occurence of infective diseases of pigs (Obremski et al , 2008; Prodanov-Radulović et al., 2011).

The aim of the paper was to evaluate the influence of mycotoxins on the health status of suckling and weaned piglets.

MATERIAL AND METHODS

The material for this research included the samples from five swine farms, where health disorders in suckling and weaned piglets were detected. Depending on the specificity of each evaluated case and available material, the applied research methods included: epidemiological and clinical evaluation, pathomorphological examination, standard laboratory testing for detection the precence of aerobic and anaerobic bacteria, virological testing and microbiological feed testing, in order to examine the presence of fungi and mycotoxins by the method of thin layer chromatography.

RESULTS AND DISCUSSION

In the first examined farm, the health disturbances in suckling piglets and weaners were registrated. Clinically, the diarrhoea in suckling piglets already in the first 3 days of life after farrowing were detected. After supervision of the farm records several facts were discovered: diarrhoea occurs in the piglets of normal birth body weight, the percent of mortality is higher in animals in good body condition and on the weaning there is 30% of small piglets. Therapeutic treatment of piglets by oral and parenteral antibiotics aplication did not improve health problems. On the farm dams are twice vaccinated during gestation with the aim to prevent disease in piglets (diarrhoea) in the first days of life. By clinical examination the certain number of suckling piglets the clinical sign of vulvovaginitis (swelling and reddening of the vulva) were dicovered. Carriyng health control in the weaned piglets the diarrhoea and signs of pneumonia (cough, nasal secretion) were detected. The pathomorphological examination of the dead suckling piglets revealed lesions dominantly on the mucosal surface of the digestive tract (*Haemorrhagiae mucosae ventriculi, Enteritis catharralis acuta*). In dead weaners beside lesions in the digestive tract, the prominent pathological changes in lungs were discovered (*Pneumonia fibrinosa in statu hepatisationis rubrae et griseae*). By bacteriological testing on tissue samples deriving from dead animals the following bacteria was detected: *Escherichia coli haemolytica, Streptoccocus alfa haemolyticus, Pasteurella sp.* Having in mind the clinical and pathological symptoms observed, especially the signs of vulvovaginitis in just farrowed piglets, a justified suspicion on the presence of mycotoxins in feed was made. Microbiological first feed for piglets testing detected 3-fold increase in the number of fungi genera *Fusarium sp, Penicillium, Aspergillus, Rhisopus sp.* as compared to the level set by the regulation. Applying further laboratory testing increase of the total number of fungi in the large number of examinated feeds was discovered: corn ($887x10^3$ *Aspergillus, Rhisopus*), piglets second feed ($319x10^3$ *Aspergillus, Mucor, Rhisopus*), feed for pregnant sows ($123x10^3$ *Penicillium, Fusarium*) and feed for lactating sows ($526x10^3$ *Aspergillus, Penicillium, Mucor*). The presence of mycotoxins was detected: zearalenon (ZEA) in the feed for pregnant sows (0.72 mg/kg), aflatoxin (AF) B1 in the piglets first feed (0,018 mg/kg) and ochratoxin (OCT) A in the piglets second feed (0.12 mg/kg).

On the second examined swine farm the health problems included increased incidence of clinical and pathomorphological signs of infective diseases. Analysing the existing data on the farm, the high mortality in piglets 7 days before weaning was noticed, which do not decrease after medical treatment. With the aim to overcome the problem, the measure of medical treatmet of piglets 3 days before weaning was introduced, but again with no result. In the weaned piglets the disease was clinically characterised with the signs of severe yellowish diarrhoea, dehidratation, huddling, roughly hair and sporadically coughing. Therapeutic treatment of the diseased animals was multiple: the antibiotics were given through feed, water and parenterally. Appying pathomorphological examination on the dead weaned piglets, the prominant changes on the digestive and respiratory tract were detected (*Gastroenterotyphlitis haemorrhagica, Poliserositis fibrinosa massiva, Pneumonia complex*).

By bacteriological testing on tissue samples from dead piglets the following bacteria were isolated: *Escherichia coli haemolytica, Pasteurella sp, Streptococcus uberis* By laboratory feed testing i. e. the available first feed for piglets the presence of OCT (0.5 mg/kg) and ZEA (4 mg/kg) was discovered.

The presence of mycotoxin in feed for pregnant sows have influence on the occurence of decreased immunological defence in piglets (Prodanov et al., 2009). As a consequence of immunosuppresive action of mycotoxins, clinical and pathological lesions correspond to the infective diseases of different ethiology (Kabak et al., 2006; Obremski et al., 2008). Combinations of several mycotoxins may potentiate the action of one other, or at least exert an additive effect (Osweiler, 2006). Nutritional effects associated with feed refusal may also contribute to observed decreased efficacy of therapeutic treatments and vaccination (Oswald et al., 2005). In the second evaluted case, the presence of OCT and ZEA in the swine feed was detected. Consequently, on the farm an evident decrease in the swine immunity against infective diseases was noticed and there was no positive respond on the applied antibiotic therapy. Also, the occurence of diarrhoea and increased percent of waste piglets can be connected with presence the mycotoxins in the feed, because the piglets display clinically feed refusal. The gastrointestinal tract represents the first barrier against ingested food contaminants and natural toxins. Following ingestion of mycotoxin contaminated feed, intestinal epithelial cells could be exposed to a high concentration of toxin (Bouhet and Oswald, 2007). From a public haelth perspective, increased infections in animals may result in increased animal-to-human transmission of pathogens and/or increased antibiotic concentrations in meat, as a consequence of animal treatment (Oswald et al., 2005).

The complex health problems on the third swine farm were noticed. Applying control of the anamnestic data, lately the frequent periods when sows delivery mummified piglets, stillbirths and decreased litter size were observed. Beside this, just farrowed piglets are nonviable and despite the medical treatment, they live only 3-4 days after birth. In the pregnant dams the immunoprophylaxis is carring out with the aim to prevent the outbreak of disease in suckling pihglets. Beside this, the antibiotics are added in the sows feed 7 days before and 7 days after farrowing. Sporadically, the occurence of severe yellowish diarrhoea in piglets and apparent clinical signs of vulvovaginitis in just born piglets were evident. In the weaners the clinical signs of diarrhoea and pneumonia are sporadically noticed. The pathomorphological examination of the dead weaners revealed lesions on the organs of respiratory (*Pleuropneumonia actinobacillosa, Pneumonia interstitialis*) and digestive tract (*Gastroenterotyphlitis haemorrhagica acuta*). Applying bacteriological examination on the tissue samples deriving from dead piglets the following bacteria were isolated: *Escherichia coli haemolytica. Pasteurella sp, Actinobacillus suis.* After laboratory testing of swine feed samples the simultaneous presence of several mycotoxins was established: ZEA (6.4 mg/kg), AF (0.0064 mg/kg), OCT-A (0.032 mg/kg).

For the known mycotoxins of clinical importance for swine production, the response is usually subacute or chronic and the presenting signs are often subtle and vague (Gonzales and Rodriguez, 2008). The continuos intake of small amounts of mycotoxins leads to chronic intoxication which is clinically characterized by the loss of weight, insufficient weight gain and increased susceptibility for infectious diseases (Osweiler, 2006). Ingestion of low doses of mycotoxins can increase intestinal colonization by opportunistic pathogenic bacteria in piglets (Oswald et al., 2005; Taranu et al., 2005). Mycotoxins mixtures i.e. the combinations of several micotoxis are likely to occur naturally and they may influence on the immunity in an additive or synergistic manner. Economic losses that occur as a consequence of interaction of several mycotoxins are still unknown because in low concentrations several mycotoxins may interact in way that is difficult to detect. Combinations of several and more moderate concentrations of different mycotoxins, which individually may appear to be too low in level to be a concern, can cause cumulative toxicosis, which affect the ability of the pigs organism to fight diseases (Diekman and Green, 1992; Osweiler, 2006).

On the fourth examined farm, clinically in suckling piglets the sings of severe distrurbance of the central nervous system (wide open eyes, paddling, trembling, ataxia, paresis and paralysis) were detected. In some cases the whole litter of piglets died within 48 hours.

Despite the fact that the piglets were therapeutically treated, there was no evident respond to applied medication. The pathomorphological changes that were detected in dead sucklings indicated the lesions characteristic for *Morbus Aujeszky* infection (MA) (*Necroses miliares hepatis, Haemorrhagiae corticis renis, Tonsillitis diphtheroides necroticans*). By microbiological testing in feed for lactating sows the precence of fungi (*Fusarium sp., Mucor*) and AF (0.02 mg/kg) were detected. Applying virological testing (viral isolation on the susceptible cell culture) from the tissues deriving from dead piglets the MA virus was isolated.

Aflatoxins (B1, B2, G1, G2) are recognized as immunomodulatore agents, and when AFB1 is metabolised by mammals occurs in milk as M1. It is assumed that AFB1 is the most toxic fraction (Osweiler, 2006; Živkov-Baloš et al., 2008). In the case where the outbreak of MA on the farm were examined, mycotoxin (AF) in the feed can be connected with the possible reactivation of chronioc (latent) infection. It has been discovered that aflatoxins decrease resistance to bacterial, fungal, viral and parasitic diseases in swine. Subsequently, vaccinations against various infective diseases may be less effective in animals exposed to mycotoxins (Diekman and Green, 1992). Even when is present in low doses, AF alters the immune response and this may predispose pigs to infectious diseases. From the epidemiological point of view, it is important that mycotoxins may cause breakdown of active immunity and occurence of disease even in properly vaccinated animals (Marin et al., 2002; Oswald et al., 2005; Taranu et al., 2005).

In the last examined case, in suckling piglets the occurence of neonatal diaorhea already in the first 3 days of life after farrowing were detected. These health problems did not improve after the medical treatmant with antibiotics. In great number of just farrowed piglets the most prominent clinical sign was vulvovaginitis (swelling and reddening of the vulva). Beside this, a large number of small, weak and splayleg piglets were noticed. The newborn piglets were weak, nonviable, with diarrhoea. The diseased piglets lived only for 4 days after birth. They probably died due to hypoglicemia, because sows did not have enough milk or the piglets were too weak and did not have enough strenght for milk suckling. Applying patomorphological examination on the dead suckling piglets the prominent changes on mucosal surfaces of the digestive tract (*Gastroenteritis haemorrhagica*), less number of pale kidneys, necrotic and distrophic processes on liver tissue were detected. By laboratory testing of the available swine feed grains, the presence of ZEA in different concentrations was detected (from 0.72 to 6.4 mg/kg). The ZEA mycotoxicosis in weaned piglets was clinically characterised with signs of pneumonia, slow growth, vulvovaginitis and necrosis of the tails, sporadically with diarrhoea and rectal prolapses. The patomorphological examination of the dead weaners revealed the following lesions: bleeding on the mucosal surface of the digestive tract, pleuropneumonia and pneumonia, hepatomegalia, focal nephritis and rectal prolapses. Ethiologically the pneumonia was caused by *Actinobacillus pleuropneumoniae, Haemophilus suis, and Mycoplasma hyopneumoniae*. Also the problem that was frequently observed was digestive infection with enteropathogenic *Escherichia coli*, which can be potentiated with the detected mycotoxin (ZEA 0.8 mg/kg) and a high number of different fungi species in weaners feed (*Fusarium, Penicillium, Aspergillus, Rhisopus*).

Zearalenone is a mycotoxin which as an estrogen binds competitively to estrogen receptors of the uterus, mammary gland, liver and hypothalamus (Gajecki, 2002). Pigs are the most prone to the presence and negative effects of ZEA (Diekman and Green, 1992; Obremski et al., 2003). In our research, the perinatal hyperestrogenic syndrome was a constant clinical sign in suckling piglets. This is certainly the consequence of mycotoxins presence in feed for sows, e.i. during the pregnancy and the presence of its excreted metabolite in milk of the exposed sows.

CONCLUSIONS

The influence of mycotoxins on immune system is of special interest in swine industry. The technology on swine farms demands frequent vaccinations, especially in piglets and sows which may be a problem in the case of immunocompromised animal. From the obtained

results an example of immunosuppresive effect can be presented i.e. the occurence of enteroxemia in piglets, despite the fact that dams were vaccinated twice during gestation. The enterotoxemia is caused by pathogenic bacterial strains and occurs frequently as a cause of mortality in the young categories. It can be provoked with the feed quality i. e. the presence of mycotoxins.

The presence of mycotoxins in feed can be directly connected to the detected health disturbances of the examined piglets. In our research we discovered the persistant presence of various infections, which react poorly or do not react on the applied antimicrobial therapy (gastroenteritis, pneumonia). Also, the chronic disturbances, for instance slow growth, malnutrition, vulvovaginitis suggest on the potential presence of mycotoxins.

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