

## Disposal of Animal Waste as a Risk Factor in the Spread of Zoonotic Pathogens

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### Abstract

The aim of this study is to give an overview of the production and management of animal waste in the Republic of Serbia, and to emphasize the importance of this issue and its proper solution. Disposal of animal waste is a significant prophylactic measure, aimed at detecting, preventing, suppressing and eradicating infectious and parasitic diseases in humans and animals. The process of animal waste disposal starts from the manufacturer, who is responsible for ensuring safe storage of waste in a cooling unit, until its final removal in the processing plant. Disposal of animal waste must be done in a safe way otherwise dangerous pathogens can be spread by water, air and soil and can endanger the health of the population. In the European Union, and the Republic of Serbia accordingly, the legislation was introduced regulating the ways of managing animal waste, categorizing hazardous waste and ways to its secure resolving, with member states obliged to respect and apply the methods of safe handling of by-products of animal origin.

*Key words:* animal waste, zoonotic pathogens, pathogen survival, safe disposal

## Introduction

Animal waste is one of the most complex environmental problems nowadays. Particular importance lies in its potential dangers in terms of pollution of basic spheres of life: water, air and land (Plavša et al., 2011). Animal waste is an organic matter that originates from livestock production (carcasses, killed animals, meat and milk processing waste, rawhide processing waste and waste produced by laboratories in diagnostics of infectious and other animal diseases, laboratories for in vivo experiments, products of animal metabolism in the form of liquid and solid manure). Animal waste gets into all spheres of life in different ways and is potentially a threat to human and animal health. In order to protect the health of people and animals, all livestock producers, regardless of the size of their farms, have an important role in the restriction of movement of pathogenic microorganisms in the environment. Therefore, the management of waste generated by livestock farms as well as by the facilities for animal products processing, is very important and must be directed to improvement of animals housing, modification of animal nutrition, improvement of security of production facilities (facilities for processing of meat and milk, rawhide processing etc.), a safe disposal of animal carcasses and animal slaughtering by-products proper disposal of manure as well as its proper chemical and biological treatment. The number and type of microorganisms in animal waste depends on animal species, type of feed and feeding method, animal health status, manure characteristics and ways of its disposal.

### Condition of Animal Waste in Serbia

Animal waste management includes collection, sorting according to risks (Categories 1, 2 and 3) and the following treatment for safe disposal. Animal waste is classified into the waste group 02 00 00 according to the Waste Catalogue. According to the data given in the Waste Management Strategy (2010) in the Republic of Serbia, 900 slaughtering and meat processing plants were registered, and over 260 000 cattle, over 1 655 000 pigs and more than 2 300 000 poultry and other animal species heads are grown. The annual production of animal waste in Serbia (carcasses and slaughter confiscates) is about 28 000 tons of carcasses and 245 000 tons of slaughterhouse waste, out of which only 20% is processed in rendering plants. The rest is usually dumped without treatment in landfills and buried. There are open type installations for treatment of animal waste in Sombor (Categories 1 and 2), Belgrade (not functioning), Čuprija (Categories 1 and 2), Bačka Topola (Category 1), and several closed type processing plants (in Žitište, Plandište, Sremska Mitrovica, Sjenica, Vrbas).

In Indija, Serbia, there is another state of the art factory "Energ zelená", built for processing of Category 1 animal waste, with a capacity of about 150 000 t/year, which is also out of function. As for the quantity of manure in Serbia, according to available sources, medium and large livestock farms (cattle, pigs and poultry) produce about 10,310 m<sup>3</sup> of manure on a daily basis. According to the data provided in the Waste Management Strategy 2010-2019 (Official Gazette of RS 29/10), the quantity of animal waste amounted to 277 000 tons in 2010, 296 000 tons in 2014, and projections for 2019 are as much as 321 000 tons.

Generated animal waste contains various forms and types of microorganisms that can cause occurrence of zoonoses and spreading of infectious diseases among animals. This material gets into natural resources and, if not safely removed, poses a threat to land, water and air and endangers the public health of the population and livestock. Pathogens of bacterial etiology (*Salmonella sp.*, *Campylobacter sp.*, *E. coli O157: H7*, *Listeria*, *Yersinia enterocolitica*, *Anthrax*, *Leptospira*, *Coxiella burnetii* etc.), viral (*Rheo* viruses, *Rota* viruses, *Pesti* viruses, *Coxsackie* viruses, *Adeno* viruses, etc.), and parasitic protozoa (*Cryptosporidium parvum* and *Giardia Lambli*) contained in waste materials, can stay vital and infectious for a relatively long time. The aerobic microorganisms survive longer in the tissues of dead animals, if carcasses are not removed safely. Poor management of animal waste opens up the possibility of various infections via natural vectors such as insects, rodents, dogs, wild beasts, birds and others.

### Dangers Related to Animal Waste

Improper handling of animal waste threatens the basic living conditions in ecosystem, such as water, air and soil. Pathogens can survive in the external environment from few days up to several months (*Bacillus anthracis* lives in soil for several decades), depending on a type of pathogens, ecological conditions, as well as chemical, biological and physical composition of the environment (Plavša et al., 2012b). Inactivation of bacteria, viruses and protozoa in soil, water, manure, crop and livestock products, can be conditioned by temperature, pH value, UV radiation, inorganic ammonia, organic nutrients, osmotic pressure and competition. The importance of each of aforementioned factors is closely linked to the environment. An environment rich in nutrients, such as animal waste, protects microorganisms from drying, temperature fluctuations, solar UV radiation, thus allowing their growth and development. Adverse living conditions such as extreme temperatures, high evaporation and high humidity and insufficient aeration significantly shorten the life of microorganisms.

Studies have confirmed that a large number of bacteria survive significantly long in the waste of animal origin such as carcasses, by-products, solid or liquid manure or other sources. Thus, *E. coli O157: H7* survives in feces for 42-84 days at a room temperature, and at lower temperatures it can survive up to 160 days, in slurry at a temperature of 20° C it lives for 2 days (Mitscherlich, 1984). Nicholson et al. (2002) tested the survival of *E. coli O157: H7*, *Salmonella*, *Listeria* and *Campylobacter* in cattle, swine, and poultry manure at 40-60° C and found that solid manure aeration reduces the survival of *E. coli O157: H7* and *Salmonella* by as much as 88%. *Salmonella* survives in feces from 96 to 190 days at a room temperature and in the liquid manure at a temperature of 10° C it survives up to 140 days.

Kudva et al. (1998) confirmed similar changes in *E. coli O157: H7* in sheep manure, where this pathogen survived 630 days at a temperature below 23° C without aeration, and 120 days when the manure aeration was performed. *Bacillus anthracis* spores are highly resistant to physical, chemical and biological treatments and are able to survive for a long period of time (tens of years) in the environment (soil). Bacteria of *Brucella* species can cause abortions and other causes of infertility in cattle, pigs, poultry, goats, sheep and dogs, being ejected from sick animals through the excretion of urine, feces, milk, seeds, aborted fetuses and others, thus getting into manure. Their survival was confirmed in cow manure up to 122 days at a room temperature, while at the temperature of 25°C they can survive for 10 days (Mitscherlich, 1984).

Also, the bacteria of the *Campylobacter* group are present in a high percentage in the manure and feces of infected animals. Skirro (1998) found 3.5% of positive samples tested for *Campylobacter* in cattle and even 59% of positive samples in poultry. The survival of these bacteria is greatest at low temperatures, so the refrigerator meat has a high risk of being a source of infection, especially poultry meat. Survival period of *Campylobacter* at 4° C is 3 weeks in feces, 4 weeks in water and 5 weeks in urine (Blaser, 1980). *Campylobacter* is unable to survive within the pH value 1-4, and at temperatures exceeding 47° C (Ebigwei, 1993).

Clostridia spores (*Clostridium perfringens*, *C. botulinum* and *C. tetani*) are very durable and resistant to various physical, chemical and biological treatments and can survive in the soil, in the external environment, for many years (up to a few decades). The concentration of *C. perfringens* in feces and manure of farm animals is relatively high and can be up to 10.000 org./g (Hutchison et al., 2005). The zoonotic pathogen *Chlamydia psittaci* survives up to 11 days in feces and urine, for more than 60 days in bird's food at room temperature, and for more than 50 days in frozen meat (Mitscherlich, 1984).

*Coxiella burnetii*, causative agent of Q-fever, survives 7-10 months at a temperature of 15-20° C on a surface, 1 month in fresh meat, 40 months in skimmed milk at room temperature, and 586 days in dry faeces (Mitscherlich, 1984).

*Erysipelotrix rhusiopathie* survives 37 days in manure at 35-37° C and more than 180 days at a temperature between 2 and 5° C (Mitscherlich, 1984). *Francisella tularensis* is very resistant and survives for several weeks at low temperatures in water, soil, hay, straw and old animal carcasses. It survives 5 months in water at 4° C, 23-35 days in contaminated water kept at 7° C, and at 21- 24° C it can survive 12 days. *Leptospira sp.* most commonly enters a manure via infected urine or placenta, and does not survive the drying of the area, low temperatures, exposure to heavy metals other than iron and if the pH is lower than 6.8 (Mitscherlich, 1984). *Listeria monocytogenes* survives 12 days in soil exposed to the sun, whilst remaining infective even up to 182 days in the sheltered soil. In cattle feces it survives 182-2190 days at a temperature of 50° C, and in liquid manure it survives 36 days in summer and 106 days in winter (Ryser, 1991).

Parasitic protozoa such as *Cryptosporidium parvum* and *Giardia lamblia*, survive in the animal manure very differently, depending on temperature conditions. *Cryptosporidium* survives 1 hour at -70° C, 1 day at -20° C, 1 or more years at 4° C, 3 - 4 months at 25° C, 1 - 2 weeks at 35° C and only 1 minute at 64° C (Fayer and Nerad, 1996; Finstein, 2004). *Giardia* cysts have very similar behavior, being only slightly less resistant to temperature extremes.

Information about the survival of zoonotic viruses (*Picornavirus*, *Rotavirus*, *Parvovirus* and *Adenovirus*) is rare in animal waste. Keretnyi et al. (1999) confirmed that swine hepatitis E virus retains in the sample of swine excrete for over 2 weeks regardless of whether the samples were held at -85° C, at 4° C or at room temperature. Pesaro et al. (1995) confirmed that viral pathogens remain much longer in manure and that they deserve more research and attention in order to prevent the spread of these pathogens in the environment inhabited by people and animals, whether domestic or wild. Wild animals, being not under full human control, are very often a source of infectious pathogens and an important link in zoonotic diseases spreading chain.

Pathogenic micro-organisms remain in water for a long time and thus are easily spreading and being transferred. Some microorganisms can remain in water and still be capable of new infection even after 448 days (*Yersinia enterocolitica*). *E. coli O157: H7*, zoonotic pathogen that is very commonly found in epidemiological reports, survives even 90 days in drinking water and over 300 days in surface waters.

Water pollution is a significant environmental problem, and it is considered that clean rivers are almost nonexistent, and a tendency is that water will soon become more demanded than oil, with a forecast that about a third of mankind will suffer of thirst even before 2025. At the International Conference on Water, Paris, 1998, it was noted that the shortage of water would be the most critical factor that might endanger the society (Pantelić et al., 2006).

Air pollution with zoonotic micro-organisms can have a great impact on spreading of airborne infections such as tuberculosis, Q-fever and others. This is stressing out the enormous importance of zoohygienic conditions in animal facilities, as well as of safe disposal of animal waste. Intensification of livestock production drastically increases concentration of toxic gases. Although the greatest generators of gases are electricity plants, producing up to 65% of total SO<sub>2</sub> emissions in the USA, livestock and crop production are also very important sources of toxic gases. Pollution of surface and deeper layers of soil often occurs due to improper disposal of carcasses, liquid and solid manure from farms. The pathogens like *Salmonella* and *Campylobacter* can survive up to 120 days, *E. coli O157: H7* up to 34 days, and *Listeria* up to 128 days. Particularly interesting is the slurry from swine farms, where *Salmonella* can survive even up to 299 days. The survival ability of those zoonotic pathogens should indicate a necessity for more stringent measures in combating and eradicating highly dangerous infectious diseases often transmitted from animal to animal, but also from animals to humans, causing frequently minor or major epidemics which sometimes end up lethally.

Great dangers are the animals that died of diseases whose causative agents produce spores (e.g., anthrax, blackleg). Sporogenous forms are very resistant to environmental factors and their survival expectancy is very long. In moist soil, *Bacillus anthracis* spores survive from 6 to 30 years. Brucellas can survive 3 to 4 months in the soil and the virus of New Castle disease survives up to 7 weeks in the carcass, whilst *Mycobacterium bovis* survives up to 167 days buried in cattle lungs, and in the intestinal tract even over 187 days.

Disposal of animal waste produced in livestock production, be it carcasses, their remains, liquid or solid manure, requires a lot of attention and a proper disposal procedure, to minimize spreading of zoonotic pathogens (Orlić et al., 2005; Plavša et al., 2005; Delić-Jović et al., 2012). The process of disposal of animal waste starts from the manufacturer, who is required to ensure safe storage of the waste in a cooling unit till the final collection and its removal in the treatment facilities. The time of disposal of carcasses and other animal waste sources is very important and therefore it is recommended for carcasses to be disposed of as soon as possible from the place of origin, within 12 hours during hot weather periods and within 24 hours during winter period.

Processing of animal waste of categories 1 and 2 provides the finished products like technical fat and meat and bone meal, to be used for technical purposes as energy fuels. Usability rate of this material is 10-15% for technical fat and 20-25% for meat and bone meal. One of the next steps in the establishment of management and control system of safe disposal of animal waste, is the creation of a register of polluters and determining of the volume of animal waste production, mandatory sorting at the place of origin, proper labeling and storage in cooling units, until final disposal, to preserve the quality of organic matter and to prevent spreading of pathogenic microorganisms (Košarčić et al., 2008, 2009).

The European Union (EU), and the Republic of Serbia accordingly, introduced the legislation governing animal waste management methods. The EU adopted Directive 999/2001, stipulating the rules on prevention, control and eradication of Transmissible Spongiform Encephalopathy and Directive 1774/2002, and its Annexes, on categorization of hazardous waste and methods of safe sanitation, obliging the member states to respect and apply the methods of safe treatment of animal origin by-products. In October 2009, a new EC Regulation No. 1069 was brought, defining sanitary rules regarding animal by-products and derived products not intended for human consumption. On the basis of European legislation, the Republic of Serbia adopted the Law on Waste Management ("Off. Gazette of RS" Nos. 36/2009 and 88/2010), and in 2011 adopted the Rules on Method of Classification and Treatment of the By-products of Animal Origin, Veterinary-Sanitary Conditions for the Construction of Facilities for Collection, Processing and Disposal of By-products of Animal Origin, Methods of Official Control and Self-control, as well as on Conditions for Livestock Cemeteries and Landfills ("Off. Gazette of RS", No. 31/2011). On two occasions, amendments were made to the Rules ("Off. Gazette of RS", Nos. 97/2013 and 15/15).

### Methods of Safe Disposal of Animal Waste

In compliance with the above mentioned legislation, methods of safe sanitation of animal waste, with respect to their classification (category 1, 2 and 3) are burning in special furnaces at temperatures of 850-1200°C, taking into account regular monitoring of gas emission limit values and concentration of dioxins; processing in rendering plants, to get economically valuable products (meat and bone meal, technical fat and hot water); and burial of animals at animal cemeteries or landfills. Considering possibilities of safe disposal of animal waste generated as a product of animal metabolism such as liquid and solid manure, there are two methods: biological (aerobic storage, composting and anaerobic digestion) and chemical treatment (chlorine, ozone, UV radiation and pasteurization) of manure (Plavša et al., 2012a).

Disposal of animal by-products and derived products should take place in accordance with the regulations of environmental protection, especially when it comes to landfills and waste incineration (Plavša et al., 2009). Waste incineration should be carried out in accordance with Directive 2000/76/EC. Conditions in respect of the operation of these plants are related to the permitted gas emission (limit values), waste water and rest (ashes) that are subject to mandatory monitoring. Burial and incineration of by-products, in particular of carcasses, may be justified in certain situations, especially in remote areas, to control certain infectious diseases when it is necessary to immediately remove killed animals/carcasses or their parts in order to prevent the outbreak and spreading of transmittable diseases. Natural disasters (floods, fires) can also induce such a situation. According to Directive 1069/2009/EU:

1. Disposal and Use of Category 1 Waste shall be carried out as follows:

- Incineration in special furnaces,
- Sterilization of waste under the following conditions (the size of chopped pieces up to 50 cm max., sterilization at the temperature of 133°C for at least 20 minutes and under a pressure of at least 3 bars, after which the waste shall be permanently marked and buried at special permitted landfills or used as energy fuel for combustion (in cement plants, etc.)

2. Disposal and Use of Category 2 Waste shall be carried out as follows:

- Incineration in special furnaces,
- Sterilization of waste under the following conditions (the size of chopped pieces up to 50 cm max., sterilization at a temperature of 133°C for at least 20 minutes and under a pressure of at least 3 bars, or can be used in the production of organic fertilizers or as a material in the process of composting and biogas production).

3. Disposal and Use of Category 3 Waste shall be carried out as follows:

- Disposal as waste for incineration, with or without prior processing,
- Recycling or disposal by incineration,
- Disposal at authorized landfills after processing,  
Mandatory treatment if the material is in a process of decomposition and decay, in order to avoid the risks of endangering public health and animal health that might be affected by this product,
- For the production of food for animals bred for fur production,
- For the production of pet food,
- For the production of organic fertilizers and soil improvement,
- As a material for composting and biogas production, etc.



According to the same directive EC 1069/2009, the Category 2 material may be determined to specific nutritional purposes, provided that there is no risk to public health and animal health under the condition that it comes from animals which were not killed or died from infectious diseases that endanger the health of humans and animals. The Category 3 material may be used for the feeding of animals (animals in zoos and circuses, prey birds, fur animals, wild animals, dogs and cats in shelters, worms and fishing worms), and as biomass for energy production (Vorkapić et al., 2012).

## Conclusion

Intensive livestock production generates an increased amount of animal waste, which, if inadequately disposed of, may represent a source of infection for humans, domestic and wild animals. This waste is also an important factor of balance in the natural environment. Animal carcasses, manure, slaughterhouse waste and other kinds of animal waste represent a specific waste material and due to the specificity they need to be treated taking care of hygienic, epidemiological, ecological and economic aspects. They should always be seen as a potentially hazardous material because they contain microorganisms of which some are zoonotic. The management of animal waste is crucial in maintaining a safe and sustainable environment, and there is an urgent need to form a Register of Polluters and to determine the volume of production of animal waste, its mandatory classification at the place of origin, proper labeling and storage in safe refrigerated chambers until its final disposal in order to preserve the quality of organic matter and to prevent spreading of pathogenic microorganisms.

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## Сажетак

Циљ рада је дати преглед производње и управљања анималним отпадом у Републици Србији, те указати на значај ове проблематике и њеног правилног рјешавања. Збрињавање анималног отпада је значајна профилактичка мјера усмјерена на откривање, спријечавање појаве, сузбијање и искорјењивање заразних и паразитских обољења људи и животиња. Процес збрињавања анималног отпада почиње од самог произвођача, који је дужан да осигура безбједно чување отпада у расхладним коморама до коначног сабирања и одвожења у објекте за прераду. Збрињавање анималног отпада мора се вршити на безбједан начин, у противном се опасни патогени шире преко воде, ваздуха и земљишта, при чему могу угрозити здравље становништва. У Европској Унији, а сходно томе и у Републици Србији, уведене су законске регулативе које уређују начине управљања анималним отпадом, категоризацију опасног отпада и начине безбједног санирања, при чему су земље чланице обавезне да поштују и примјењују методе безбједног поступања са нуспроизводима анималног поријекла.

*Кључне ријечи:* анимални отпад, зоонозни патогени, преживљавање патогена, безбједно збрињавање

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