

Biocoenosis of Construction Objects of Livestock Breeding Complexes with Due Account for Industrial Profile

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Abstract— The work studies and identifies testing cultures of microorganisms colonizing the surface of construction structures (walls and floors) exploited in natural conditions of largest agricultural cattle breeding, hog breeding and poultry (chicken and turkey) breeding sites in Belgorod region.

Keywords—biocorrosion; bacteria; micromycetes, agroindustrial complexes, sanitation

I. INTRODUCTION

The agricultural sector is the most dynamic branch of the world economy and remains the most contributing source of the world's food stock. The development of agroindustry, in particular all livestock breeding branches, is accompanied by the formation of large amount of waste [1–5]. Organic waste (excreta and manure) is an inherent part of the production process. Livestock excreta contains technological and flush water, waste of food and gases, organic acids, mineral compounds, carbon dioxide, hydrogen sulfide and ammonia, which promotes the formation of microcenosis breeding grounds [6–10].

The main tasks of agricultural industry to complete are: increased quality of sanitation and disinfection of premises; application of disinfectants with both bactericidal and fungicidal properties, because by today mycotoxicoses have taken a prominent position among diseases; careful analysis of food to identify and measure the most spread mycotoxins and search for effective agents for early diagnosis, treatment and prevention of mycotoxicoses; reduced levels of production premises contamination by microorganisms, etc.

Currently, the problem of biodegradation of the premises of agricultural enterprises is solved by sanitation and prevention measures. This process includes a whole bunch of different periodic activities: sanitation of premises and territories around, as well as preventive maintenance of premises and equipment.

To determine the efficacy of the sanitation and prevention activities at largest agricultural cattle, hog and poultry (chicken and turkey) breeding industries of Belgorod region, an inspection was run to check the presence of the indicators of biological wear of surfaces (walls and floors) of breeding premises.

II. MATERIALS AND METHODS

The sampling of the mycobiota on the sections of walls and floors of the premises at livestock breeding complexes and poultry farms was performed by rinse sampling. The washings were taken from both horizontal (floor) and vertical surfaces (walls) with the area of 100 cm² (Fig. 1); the rinsing of cultures was made between sanitations, several days before the next sanitation of premises. The sampling was made by sterile cotton swabs made of viscose (Fig. 2) put into a vial with 2 ml of sterile solution of NaCl. Because the access to the enterprises was restricted, the test subjects were sampled by the specialists of enterprises of corresponding profile.

In the laboratory, the samples were diluted by sterile solution of NaCl to the degree that corresponded to the bacterization of the surfaces under study: 1 to 10, 1 to 100 and 1 to 1000.

The bacterial content was determined in meat infusion agar; that of mycelial fungi was determined in Czapek-Dox medium. Into each Petri dish, 20 ml of medium was poured and sent for sterilization. Before the study, the dishes with medium were thermostatted for 1 day at a temperature from 30 to 35 °C to confirm medium sterility.



Fig. 1. Process of rinsing from floor and walls



Fig. 2. Sterile cotton swabs used for sampling

After careful mixing of vial contents with washings, inoculation was performed on agarized media using Drigalski spatula. Then, the Petri dishes were put into the thermostat for incubation at a temperature from 20 to 25 °C for 72 hours. After incubation, the number of colonies was determined in three parallel dishes; they were smeared, fixed and Gram-stained and microscopied to identify the microorganisms.

III. RESULTS

The analysis allows concluding that the agricultural premises are populated by both bacteria (Table I) and mycelial fungi (Table II).

In the animal enclosure premises, the surfaces of walls and floors contain *Bacillus* bacteria, which are evidently the most adapted to aggressive environmental factors.

The action of the bacteria is not evident; however, their effect can be appreciably higher than that of fungi. In certain cases their presence is hard to detect with a naked eye.

The damage can manifest as foreign odor, stains, slime, etc. However, the mechanism of their action is similar to mold fungi: cracking, loss of strength, growth of potentially pathogenic and pathogenic bacteria in microbiocenosis. Because bacteria can adapt to materials with time due to their deterioration and fouling and their amount can grow, various enzymes and metabolic products can appreciably intensify corrosion of the materials. Nevertheless, the rate and level of impact depend on the schedule and type of disinfection of the

premises. In this connection, there can be no definite conclusions on the probability of corrosion effects.

TABLE I. SPECIES DIVERSITY OF BACTERIAL MEDIUM DEPENDING ON FARM TYPE

Bacteria name	Farm type			
	Cattle breeding	Hog breeding	Poultry breeding	
			Chic ken	Turkey
<i>Bacillus</i>	+	+	+	+
<i>Staphylococcus</i> spp.			+	+
<i>Streptococcus</i> spp.				+
<i>Proteus</i> spp				+

The most frequently, the animal breeding premises are populated by *Penicillium*, *Aspergillus* and *Mucor* fungi (Table II).

TABLE II. SPECIES DIVERSITY OF MYCELIAL FUNGI DEPENDING ON FARM TYPE

Bacteria name	Farm type			
	Cattle breeding	Hog breeding	Poultry breeding	
			Chic ken	Turkey
<i>Aspergillus niger</i>	+	+	+	+
<i>Aspergillus</i> spp.			+	
<i>Mucor</i> spp.		+		+
<i>Penicillium</i> spp.		+		+
<i>Cladosporium</i> spp.	+		+	

Mold fungi cause chemical (by metabolites) and mechanical damage (fouling and penetration by mycelium hyphae into a material) of materials. The main chemical products of fungi metabolism causing damage of materials are extracellular enzymes and organic acids.

In addition to purely chemical destruction of materials, microorganisms and metabolites can change their physicochemical properties due to cracking. Biofouling can lead to deterioration of decorative and other external properties of the materials, e.g. mold stains; though the functionality of the object can preserve, they can be a source of spores in the air of the premises, which can lead to diseases and increased loss of cattle.

Despite the similarity of populated microorganisms, the species diversity of corrosion agents is characterized by certain discrepancies. The samples cultured from washings taken from cattle breeding enterprises predominantly contain gram-positive bacteria (Fig. 3).

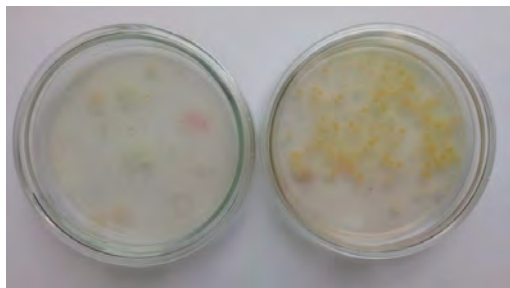


Fig. 3. Bacteria colonies in dense nutrition medium

For instance, we have detected smears of *Bacillus subtilis* and *Sarcina* (Fig. 4). *Sarcina* bacteria (Fig. 4b) are fairly heterogeneous; they can be found in water, soil, living organisms, on the surface of grass seeds. They decompose organic residues containing proteins and form ammonium salts. After division they form “bags”; their formation depends on environment. They are gram-positive saprophytes, do not form spores and can form capsules.

Bacillus subtilis is a movable rodlike bacterium (Fig. 4a) with the length of about 5 μm and diameter of 0.7 μm . It is aerobic bacterium which is widely spread in soil and vegetable residues. It decomposes organic substances and often causes food spoilage, including animal food.

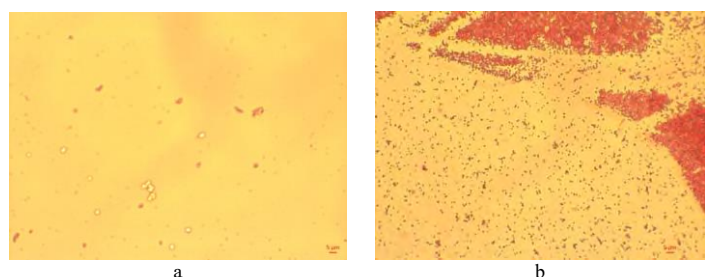


Fig. 4. Bacteria detected in washings: a – *Bacillus subtilis*; b – *Sarcina*

It forms spores (Fig. 5), which enables it to preserve in environment for a long time. High biological activity is its special feature. *Bacillus subtilis* is very mutable which hinders its identification using cultural properties.

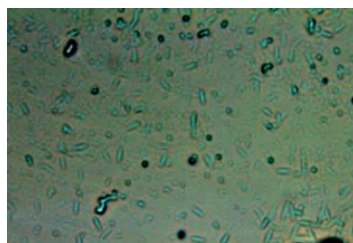


Fig. 5. *Bacillus subtilis* dyed by methylene blue

The intensity of growth and development of microorganism culture depends on the rational relation of mineral and organic substances in the environment. The spore formation depends on nutrient sources; the spores form on poor nutrient media and in unfavorable environment.

Bacillus subtilis are antagonists of many pathogenic and potentially pathogenic bacteria (they release antibiotics); besides, they acidify environment, i.e. change pH of the substrate surface on which they live, thus giving rise to the possible corrosion of construction materials.

In addition to the bacteria on the floor and walls, microscopic mold fungi were detected (Fig. 6). Cultured microscopic fungus is *Aspergillus niger*. The identified *Aspergillus* fungi are widely spread in nature and are found in soil, hay, grain, dust and animal hair. They can be brought with food.

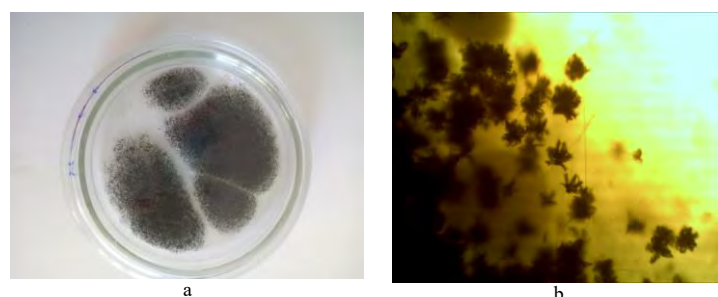


Fig. 6. Mycelial fungus *Aspergillus niger* detected on washings from surface of construction materials: a – colonies, b – sporangia

However, large concentrations of *Aspergillus* spores in air can cause diseases of both animals and human, such as lung aspergilloma. They can easily get through rhinopharynx into trachea and invade bronchi, alveoli and bronchia. After attaching they start actively developing. Toxins released by the colony promote formation of necrosis sites. As a consequence, parenchymatous tissue of lungs stops functioning leading to asphyxiation and death.

Bacterial agents in the washings of the hog breeding facility are similar to those from cattle breeding premises. However, the mycelial fungi are somewhat different. Among the cultured fungi, *Mucor* spp and *Cladosporium* spp species were detected (Fig. 7) that belong to saprophytes.

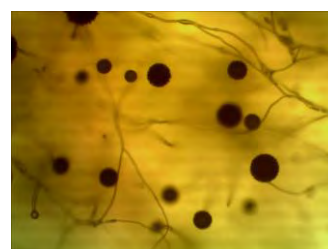


Fig. 7. *Mucor* spp fungus found in washings of hog breeding premises

In the environment they grow on decaying organic waste or compost and can be phytopathogenic.

We have also detected *Penicillium* spp fungus (Fig. 8). These micromycetes belong to cosmopolitan organisms; they are found in the same places as *Aspergillus* spp. Some of the fungi are phytopathogenic, all of them are present in soil and live on organic substrata. As a rule, they are brought with food for hogs.

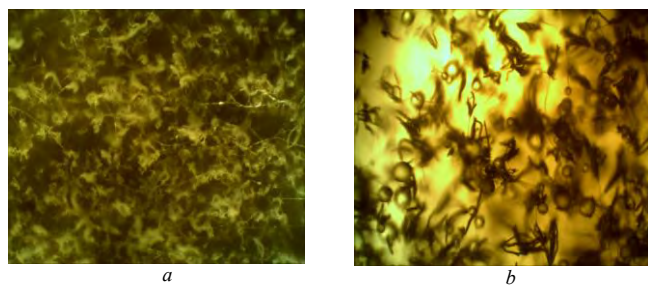


Fig. 8. *Penicillium* fungi colonies a, b

The bacterial medium of the samples rinsed from the surface of materials from the poultry breeding premises predominantly contained (as in other cases) gram-positive bacteria; however, in the total volume, gram-negative species were also found (Fig. 9).

Among microscopic fungi cultured from the floor samples, *Aspergillus spp* were detected (Fig. 10).

The *Aspergillus* fungi are widely spread in nature and are found in soil, hay, grain, dust and poultry plumage. The spores of *Aspergillus spp* can cause different diseases in both human and animals.

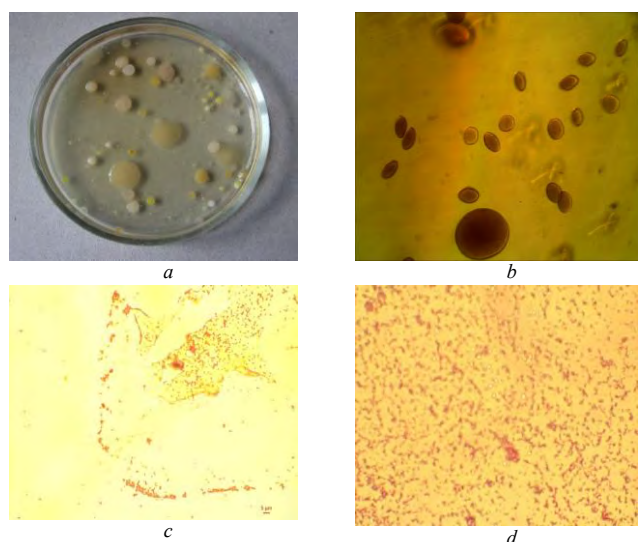


Fig. 9. Bacteria from poultry farm washings: a – appearance of nutrition medium; b – colonies; c – gram-negative *Proteus spp* bacteria; d – gram-positive *Staphylococcus spp* bacteria

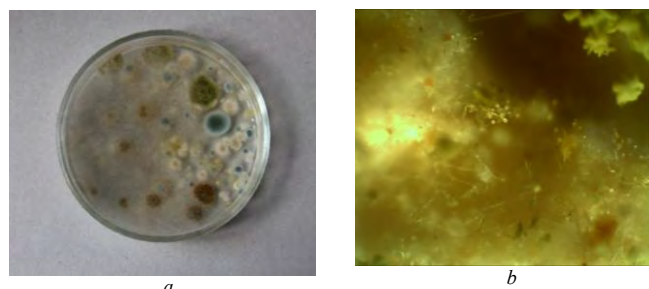


Fig. 10. Fungi cultured from the washings from the surface of construction materials of poultry breeding premises: a – appearance of colonies on nutrition medium; b – mycelial fungi *Aspergillus spp*

The study of the peculiarities of biocoenosis of turkey breeding premises was made in two ways: on the farms for breeding young (younger than 4 months) and adult (from 4 to 12 months) species. This is connected with the difference in breeding conditions depending on their age and frequency of sanitation of premises as a factor of biocoenosis resistance formed in the process of farm operation.

Among the bacteria identified in the premises with young poultry, the dominating ones were gram-positive bacteria (Fig. 11). Among microscopic fungi the following were identified: *Aspergillus*; *Mucor*; *Penicillium*.

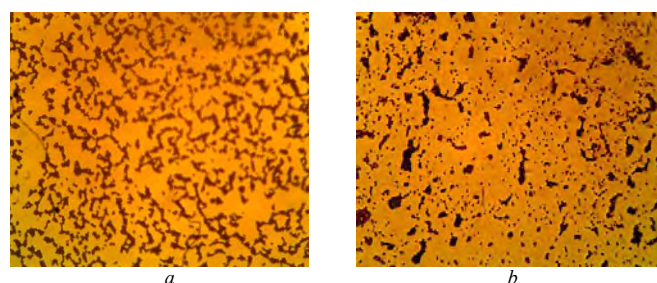


Fig. 11. Gram-positive bacteria: a – *Staphylococcus spp*, b – *Streptococcus spp*

The samples taken in premises for adult poultry, *Streptomyces actinomycetes* were found (Fig. 12). To estimate potential hazard from biocoenosis formed on the material surface (bacterial and mycelial) total bacterization of structures was calculated (Table III).

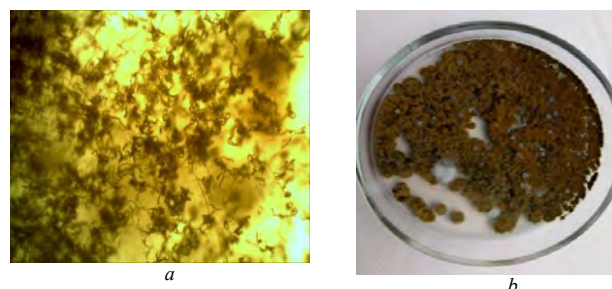


Fig. 12. The *Streptomyces* cultured from the samples taken from the construction material surface of adult turkey breeding premises: a – colonies; b – appearance of nutrition medium

The calculation of corrosion agents was made using a counter of colonies with consequent recalculation with a due consideration of the rinsing area (10 cm²).

According to the received data, total bacterization by microorganisms in the case of cattle, hog and chicken houses is almost the same. The structures of floors are more populated by both bacteria and fungi, which is primarily connected with the release of different waste during the life activities of animals and poultry.

The appreciable quantitative dominance of bacteria in turkey houses, regardless the age, is noteworthy: the concentration of bacteria is 10–100 times higher than similar indicators for the farms breeding other animals. The structure of the premises with young species is also highly populated by

mycelial fungi; on average the figures are two times higher for the floors as compared with other farms, and hundreds of times higher in the case of walls.

TABLE III. TOTAL BACTERIZATION OF CONSTRUCTION STRUCTURES OF LIVESTOCK BREEDING COMPLEXES

Farm type	Amount [pcs/cm ²]			
	Bacteria		Fungi spores	
	Wall	Floor	Wall	Floor
Cattle breeding	$0.9 \cdot 10^4$	$1.3 \cdot 10^4$	–	10
Hog breeding	$1.3 \cdot 10^4$	$1.8 \cdot 10^4$	less than 10	300
Poultry breeding				
– chicken	$1.1 \cdot 10^4$	$2.2 \cdot 10^4$	–	10
– turkey (young species)	$1.5 \cdot 10^4$	$4.8 \cdot 10^5$	480	600
– turkey (adult species)	$6.3 \cdot 10^6$	$6.9 \cdot 10^5$	300	220

The degree of surface population by microorganisms depends on the object character and its location, occupancy of the premises by animals and poultry, activity of these animals, breeding type, ventilation, air moisture, total sanitary conditions. On the walls of livestock breeding complexes, the microorganisms mostly appear after sedimentation of aerosols that form when animals cough, snort, gasp and move quickly during dry coarse food distribution and during other technological processes. On the floor surface, the microorganisms form after defecation of animals and in the case of absence or low quality of cleaning. The formed sedimented aerosols in premises gradually decay physically and biologically under different factors, which can both decrease or increase the number of microorganisms.

Nevertheless, we should note that the content of microorganisms in animal breeding premises is not high, and in the case of appropriate sanitary and hygienic (disinfection) measures cannot deteriorate animal health.

IV. SUMMARY

Thus, in the experiments we have classified biocorrosion agents causing damage and/or destruction of construction structures of breeding farms of different profile. It was established that the qualitative composition of the microorganisms cultured from different animal breeding premises is fairly similar, while the quantitative composition differs depending on the type of managed animals and poultry, peculiarities of breeding and sanitary conditions of the premises.

The test-cultures of mold fungi identified during the studies were taken from the surfaces of construction structures subjected to natural conditions of agricultural enterprises and secured as pure cultures on dense nutrition media, correctly incubated to preserve sporulation. They are to be used to estimate the funginertness and/or fungicidal capabilities of developed composite building materials with prolonged

bioresistance. Taking into regard all the above, to preserve safe regime of animal and poultry breeding and provide effective functioning of system “disinfectant–material”, one should design agricultural premises using construction materials with prolonged fungicidal and algicidal properties. This will allow for:

- increased intersanitation periods;
- transition to a new cleaning system implementing less harsh chemical compositions;
- neutralize the growth of mycotoxins at early development stages;
- prevent population of premises by microorganisms, reduce biological wear of the premises and, hence, reduce the destructive effect of the metabolism products of microorganisms.

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