Mycoplasmosis of farm animals

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Abstract. The article presents an overview characterizing the spread of Mycoplasma infections among various kinds of farm animals. It also demonstrates the urgency of the pathogen of the disease - Mycoplasma spp., its characteristics and structural features, which provide significant differences from other types of microorganisms. Most species of Mycoplasma spp. are strictly specific to the host, but some of them pose a danger to humans. Mycoplasmas in animals cause diseases ranging from acute forms of the disease to an asymptomatic course. With the development of pathology, damage to various organs and tissues is observed, which indicates a high tropism of the pathogen, which contributes to the development of a generalized form of pathology. The most susceptible is the respiratory and reproductive tract, as a result of which farms are inflicted with significant economic damage. At the same time, some of the mycoplasmas build a symbiotic relationship with the host organism. Attempts to use antimicrobial therapy, including in various combinations, do not always lead to a positive result, which is due to the development of antibiotic resistance of the pathogen. Thanks to the change in the genome, mycoplasmas have become the smallest bacteria capable of self-replication. Mycoplasmas are classified as parasites or symbionts of animals, insects and plants, while the disease itself is opportunistic. Diagnosis of the disease consists mainly of three methods: serological, molecular biological and bacteriological, which are often used simultaneously. The cultivation of mycoplasmas has its own characteristic difficulties and features due to the structure of bacteria.

1 Introduction

They were first isolated in Pasteur's laboratory in 1898, when the pathogen of cattle pleuropneumonia was identified [1]. Mycoplasmas are prokaryotic cells that are classified into a separate taxonomic group - the Mollicutes class. The most famous representatives of this class are anaeroplasmas, ureaplasma, spiroplasma, acholeplasmas. Unlike viruses, this group of microorganisms has the ability to reproduce independently, but at the same time they are devoid of a rigid cell wall unlike bacteria and are surrounded by a cytoplasmic membrane. Regardless of these features, the considered group of microorganisms is classified as bacteria. The genus Mycoplasma has about 200 species. Most of the diseases caused by the microorganism in question are chronic in animals.

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Mycoplasmas are obligate heterotrophs, which are characterized by long-term, sometimes lifelong preservation in the host organism. While in the host's body, these microorganisms produce superoxide radicals and hydrogen peroxide, which have a toxic and hemolytic effect on cell membranes and red blood cells. There are species that are dangerous both for humans and for animals and birds, for example, such as the pathogen of pneumonia in cattle and small ruminants - M. mycoides, the pathogen of rheumatoid syndrome in small cattle - M. agalactiae, the pathogen of respiratory diseases and primary atypical pneumonia in humans - M. pneumonia [2]. Mycoplasma contamination of nutrient media and cell cultures, which are used for viral replication, is of great concern [1, 3, 4].

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2 General characteristics of mycoplasmas

The pleomorphism and plasticity of mycoplasmas are the result of the absence of a cell wall; it allows them to go through the pores of filters with a diameter of 0.22-0.45 μm. A cell of the microorganism can have a spherical shape of 0.3-0.8 μm in diameter. For a long time, the question of how mycoplasmas successfully survive in various environmental conditions without a cell wall remained unresolved. It turned out that most types of mycoplasmas form biofilms. It is a well-known fact that bacteria in biofilms, as well as mycoplasmas, often exhibit a changeable phenotype compared to separately living microorganisms. Biofilms enhance the virulence, persistence and resistance of the pathogen to environmental factors. The small size of the genome determines the limitedness of their biochemical abilities, which determines their dependence on higher organisms. Mycoplasmas are characterized by gene transfer both within their class and with other bacteria.

During their evolution mycoplasmas have adapted to close interaction with eukaryotes and are parasites of mammals, fish, reptiles, arthropods and plants. Most types of mycoplasmas are opportunistic microorganisms. About a third of the known mycoplasmas are currently pathogenic to various degrees for hosts in experimental conditions or in vivo. Usually mycoplasmas start widespread reproduction and lead to pathological processes in the body with a decrease in immune forces, disruption of homeostasis. The main localizations of their habitat are mucosal surfaces of the respiratory tract, digestive system, joints and epithelium of the eyes. Most often, mycoplasmas have tissue and organ specificity. The specificity of some mycoplasmas in terms of the habitat may be due to their need for nutrients. Mycoplasmas can be transferred horizontally, autogenously, vertically, while leading to endogenous and exogenous latent course of disease. When an infectious process occurs, a focal, generalized character of the course is found [12, 15].
3 Pathogens of Mycoplasmosis in animals

In cattle the most common pathogen is Mycoplasma bovis, which provokes pneumonia among calves, mastitis, abortion, infertility and joint damage. In addition to this species, M. dispar and M. alkalescences often occurring in healthy animals, which, however, lead to pneumonia and mastitis, are widespread. M. bovigenitalium is detected mainly in the cases of the inflammation of the organs of the reproductive system, mastitis, endometritis, abortion, pneumonia and arthritis.

In pigs the disease is provoked by M. hyopneumoniae, M. hyosynoviae, and M. hyorhinis, which are detected in almost all pig farms, causing arthritis, otitis, pneumonia, etc. They pose a serious danger to humans, as they can cause malignancy. They are most often found in cell cultures, this leads to the impossibility of their further use for cultural purposes.

In birds more than 20 species of mycoplasma are found, many of which belong to commensals. Pathogenic species include: M. gallisepticum, M. synoviae, M. meleagridis, M. pullorum, M. gallopovaris, M. iners, M. iowae, M. gallinarum, M. gallinaceum. M. synoviae in turkeys and chickens causes sinusitis, arthritis, airsacculitis, and impaired egg shell formation. M. meleagridis leads to diseases of the musculoskeletal system and respiratory tract. M. gallisepticum causes chronic respiratory diseases, conjunctivitis, sinusitis, as well as a decrease in the quality of the products.

In sheep and goats, a large number of Mycoplasmosis pathogens are found, the main of which is M. agalactiae - contagious agalactia, which causes mastitis, arthritis, conjunctivitis, less often pneumonia. Pneumonia can also be caused by M. capricolum subspecies capricolum, M. putrefaciens, and M. mycoides subspecies capri. Cases of animal infections caused by M. ovipneumoniae and M. conjunctivae, which manifest themselves in the form of infectious keratoconjunctivitis, pneumonia and tracheitis, are often detected.

4 Diagnostics and cultivation of mycoplasmas invitro

Cultural, serological and molecular biological methods are used to diagnose Mycoplasmosis [14]. One of the methods of serological diagnostics is the study of smear-prints in the reaction of indirect immunofluorescence (mycoplasmas are stained bright green on the cell surface). The most sensitive method is PCR diagnostics, for which primers for the 16S and 23S rRNA genes are used. It should be noted that the use of molecular biological diagnostic methods can be difficult due to the fact that the metabolism of mycoplasma in vivo differs significantly from the metabolism of the same mycoplasma cultivated in an axenic nutrient medium [13]. The disadvantage of these methods is that if the diagnosis is confirmed by PCR or serological studies, it is impossible to select an effective antibacterial drug. Obtaining a pure culture is often necessary for the identification and selection of effective antibacterial drugs, but the limited metabolic capabilities of the pathogen make it difficult to cultivate them on artificial nutrient media.

The optimum growth temperature is 36.5-37 °C, pH 7.0. The microorganism is a facultative anaerobe. Sterols, which are an important component of cell membranes, are vital for the growth of mycoplasmas. In most cases, Mycoplasma spp. grows best in aerobic conditions, and some species in anaerobic conditions. Liquid, semi-liquid (0.3% agar) and dense (1.5% agar) media are used for cultivation, with the addition of 25% fresh yeast extract, horse whey (10-20%), meat broth. In semi-liquid nutrient media mycoplasmas grow in the direction of injection sowing, forming colonies in the form of whitish translucent lumps. On dense agar, after 24-28 hours (in some cases more) of incubation, they grow in the form of a spherical colony, which has a turbid granular center growing into the medium, with a flat openwork translucent peripheral zone. The size varies from 10 to 500 μm, on average 100 μm. On liquid media, the growth of mycoplasmas is accompanied by opalescence.
5 Virulence and pathogenicity

The main factors of pathogenicity are phospholipase, protease, cytoadhesion proteins, neuraminidase, etc. Neuraminidase, interacting with cellular structures, leads to the damage to the structure of cell membranes and the interaction of cells with each other. Proteases cleave antibody molecules and cause cell degranulation. DNase and RNase disrupt the metabolism of nucleic acids. Endopeptidases cleave IgA molecules into monomeric components [14]. Adhesins enter the surface structures, due to which the interaction by the ligand-receptor mechanism with the host cells is provided. With the help of adhesins mycoplasmas can stay in cell membranes for a long time, which protects the pathogen from antibodies, complements and other various protective factors of the macroorganism. In their turn, the metabolic products of mycoplasmas (peroxide, ammonia, carbon dioxide) have a destructive effect on the lipid membrane complexes of cells. Mycoplasma endotoxins lead to a pyrogenic effect, pulmonary edema and leukopenia [16]. In addition, penetrating into the bloodstream, mycoplasmas block the work of the mononuclear phagocyte system.

6 Antibiotic resistance and cell culture contamination

The main method of combating Mycoplasmosis and cell culture contamination is the use of antibacterial drugs. The efficiency of these measures tends to decrease, which is due to the rapid development and spread of resistance [5, 11]. Due to the absence of a cell wall, mycoplasmas are resistant to β-lactams, glycopeptides, fosfomycin and other antibiotics directly influencing the cell wall. Resistance to polymyxin, trimethoprim, linezolid, rifampicin and other antibacterial drugs is widely recorded, which is due to a mutation in the β-subunit of RNA polymerase. Antibiotics of the tetracycline and fluoroquinolone series, as well as macrolides, which are widely used in the treatment of mycoplasmas, are efficient against mycoplasmas. [6, 7, 8, 10].

7 Conclusion

Summarizing the work, it is worth making a conclusion that mycoplasmas are widespread in the world among people, animals, birds, insects, and plants. This pathogen is able to stay in the host's body for a long time, using mechanisms that allow it to evade the host's defense systems, due to its tight attachment to the cell wall and the presence of structures and substances that suppress or prevent phagocytosis.

The widespread occurrence of mycoplasmas and their role in the emergence of infectious pathology in animals create an urgent task of their rapid and reliable detection and identification of their species. The most optimal and modern method of laboratory diagnostics of mycoplasmas is the setting of a polymerase chain reaction (PCR), since the cultivation of some types of mycoplasmas is a difficult task, and the presence of cross reactions reduces the specificity of serological methods.

The solution to the problem of suppression of mycoplasmas is in the study of the mechanisms of molecular relationships between mycoplasmas and host cells. Under the influence of various factors, which suppress the body's immune system, mycoplasma may be activated. The right choice of antibiotics makes it possible to suppress the growth of Mycoplasma, but the case, in which the infection goes from acute to latent form, is not excluded. The use of inactivated vaccines does not always eliminate the pathogen in the body; animals often remain carriers.
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