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***Mycoplasma bovis*, *M. bovisgenitalium* AND *M. dispar***  
**AS BOVINE PATHOGENS: BRIEF CHARACTERIZATION**  
(review)

**M. ABED ALHUSSEN<sup>1</sup>, V.V. KIRPICHENKO<sup>2</sup>, S.P. YATSENYUK<sup>3</sup>,  
A.A. NESTEROV<sup>2</sup>, O.P. BYADOVSKAYA<sup>2</sup>, T.V. ZHBANOVA<sup>2</sup>, A.V. SPRYGIN<sup>2</sup> ✉**

<sup>1</sup>People's Friendship University of Russia (RUDN University), 6, ul. Miklukho-Maklaya, Moscow, 117198 Russia, e-mail alhussenmohammed85@hotmail.com;

<sup>2</sup>Federal Center for Animal Health Control, FGBU VNIIZh, mkr. Yurievets, Vladimir, 600901 Russia, e-mail kirpichenko@arriah.ru, nesterov@arriah.ru, bjadovskaya@arriah.ru, zhbANOVA@arriah.ru, sprygin@arriah.ru (✉ corresponding author);

<sup>3</sup>Russian State Center for Animal Feed and Drug Standardization and Quality, 5, Zvenigorodskoe sh., Moscow, 123022 Russia, e-mail pcr-lab@vgnki.ru

ORCID:

Abed Alhussen M. orcid.org/0000-0002-1210-0303

Byadovskaya O.P. orcid.org/0000-0002-8326-7151

Kirpichenko V.V. orcid.org/0000-0002-2494-3826

ZhbANOVA T.V. orcid.org/0000-0002-9857-5915

Yatsenyuk S.P. orcid.org/0000-0002-4819-2131

Sprygin A.V. orcid.org/0000-0001-5982-3675

Nesterov A.A. orcid.org/0000-0002-4288-1964

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### Abstract

The cattle mycoplasmas are widespread throughout the world (A.M. Parker et al., 2018; M. Abed Alhussen et al., 2020). This review presents data on the epidemiology and diagnosis of mycoplasmosis in cattle caused by *M. bovis*, *M. bovisgenitalium*, and *M. dispar*. Mycoplasmas can cause economically important diseases in cattle, including mastitis, arthritis, keratoconjunctivitis, otitis media, pneumonia, and reproductive disorders (R.A.J. Nicholas et al., 2008; F.P. Maunsell et al., 2011). Mycoplasmas are characterized by a size of up to 150 µm, small genome (0.58-1.38 million base pairs) a low G-C composition (23-40 %) and the absence of a cell wall which determines their polymorphism and resistance to antibiotics, influencing the synthesis of the bacterial cell wall (R.A.J. Nicholas et al., 2008; P. Vos et al., 2011). Mycoplasma surface antigens are highly variable both in vitro and in vivo, which leads to significant variability of isolates (M.A. Rasheed et al., 2017). They also play an important role in overcoming the host's immune system. In addition, some of these antigens are involved in the adhesion of mycoplasmas to host cells (Y. Guo et al., 2017). After adhesion, many mycoplasmas produce a variety of products that damage host cells and enhance pathogenesis (L.A. Khan et al., 2005). They can also form biofilms that increase resistance to drying out and heat stress (L. McAuliffe et al., 2006; F. Gomes et al., 2016). Moreover, the invasion and intracellular survival of mycoplasmas in cattle cells contributes to the preservation and spread in the host organism (J. Van der Merwe et al., 2010). The incubation period for mycoplasma infection in cattle depends on many factors, i.e., the infectious dose, the presence of associated infections, the conditions of keeping the animals in the herd and the stress state of the animals (M.J. Calcutt et al., 2018). Sick animals are a source of infection, because they can shed the pathogen with nasal discharge and sperm for several months and sometimes for several years (K.A. Clothier et al., 2010; V. Punyapornwithaya et al., 2010). It should be noted that at low temperatures, mycoplasmas remain viable for a long time: in deeply frozen cattle semen, the pathogen can remain infectious for many years (A. Kumar et al., 2011). The high contagiousness of some species of *Mycoplasma* spp., their low sensitivity to treatment and the associated consequences of culling for the affected population make timely and accurate diagnosis important for disease control and prevention (A.M. Parker et al., 2018). The cultural methods can be applied for isolation and identification of the pathogen. However, these methods have limitations. Cultivation of mycoplasmas requires a complex medium, special equipment and technical skills (R.A.J. Nicholas et al., 2008; M.J. Calcutt et al., 2018; A.M. Andersson et al., 2019). Mycoplasmas require 7-10-day cultivation at a temperature of 37 °C and 5-10 % CO<sub>2</sub>. The colony has the "Fried-egg" appearance characteristic of most mycoplasmas (P.J. Quinn et al., 2011). By contrast, PCR provides a rapid and accurate diagnosis of the disease by detecting mycoplasmal DNA (A.M. Andersson et al., 2019). Furthermore, many other methods of diagnostics of bovine mycoplasma are used, such as MALDI-TOF MS (Matrix assisted laser desorption ionization time-of-light mass spectrometry), latex agglutination,

immunochromatographic assays etc., however, each method has its advantages and disadvantages, which should be considered before application (M.J. Calcutt et al., 2018; B. Pardon et al., 2020).

Keywords: *Mycoplasma bovis*, *Mycoplasma bovisgenitalium*, *Mycoplasma dispar*, cattle, pathogens, epidemiology

Genus *Mycoplasma* (class *Mollicutes*) comprises small microorganisms characterized by the absence of cell wall, a low GC content (23–40 %) and a small genome size (0.58–1.38 million bp) [1]. The first representative of the genus *Mycoplasma*, the *Mycoplasma mycoides* subsp. *mycoides* was isolated from sick cows in 1898 and characterized as the causative agent of contagious bovine pleuropneumonia [2]. This dangerous disease, having reached a global spread in the 19th century, was successfully eradicated in most countries, with the exception of Africa, where it still remains a serious problem [3]. Other mycoplasma species that pose a threat to cattle include *M. bovis*, *M. bovisgenitalium*, *M. dispar*, *M. californicum*, *M. bovirhinis*, *M. bovoculi*, *M. leachii* (formerly *Mycoplasma* sp. *bovine* group 7) [4], *M. canis*, *M. canadense*, *M. alkalescens*, *M. arginini*, and *M. wenyonii* [5–7].

Mycoplasmas are both a secondary microflora, aggravating pathological processes caused by other microorganisms, and primary etiological agents in a host organism with a weak general resistance. Mycoplasmas cause numerous diseases in cattle, including mastitis, arthritis, keratoconjunctivitis, pneumonia, and reproductive pathologies [8]. Currently, the most common pathogenic and clinically significant species are *M. bovis*, *M. bovisgenitalium*, and *M. dispar*.

This review focuses on three pathogenic mycoplasmas of cattle, the *Mycoplasma bovis*, *M. bovisgenitalium*, and *M. dispar*, their distribution, biological properties, and laboratory methods of identification.

Clinical signs of the infections caused by *M. bovis*, *M. bovisgenitalium*, and *M. dispar*. *M. bovis* is the most common species affecting cattle, however, there have been cases of its isolation from buffaloes, small ruminants, and chickens [9]. *M. bovis* is the main cause of pneumonia in young cattle, however, adult cattle are also susceptible to this mycoplasma [10]. In calves, mortality rate due to *M. bovis* is 5–10 %, and morbidity reaches 35 % [6]. The pathogen does not cause characteristic clinical signs, and the infected animals have a sharp dry cough, fever, apathy, and discharge from the eyes (6).

When pneumonia develops, *M. bovis* is always detected in association with other pathogenic microorganisms. Lung lesions may not occur or they are limited to reddish areas of multilobular consolidation in the apical lobes [10]. However, a severe course is associated with extensive lung lesions which appear as an area of coagulation necrosis and abscesses [6]. Chronic *M. bovis* infections are characterized by lymphocytic pneumonia with hyperplasia of the peribronchial lymphoid tissue, causing stenosis of the airway lumen followed by compression and collapse of the adjacent pulmonary parenchyma [11]. The *M. bovis* antigen is found on the periphery of areas of coagulation necrosis, in necrotic exudates and is closely associated with infiltrating macrophages and neutrophils [12]. Pneumonia can occur as a single manifestation of infection or in combination with other clinical signs, including polyarthritis in adult animals and otitis media in young calves [13]. *M. bovis* can also cause subclinical, clinical or chronic mastitis, being a serious problem for milk producers [14]. *M. bovis* was also detected in aborted fetuses [14]. Both the respiratory and mastitis forms of the disease can induce arthritis [6, 16] with impaired motor function and, in severe cases, a decreased feed intake and exhaustion [6]. In addition, *M. bovis* can cause epididymitis, orchitis, urethritis, and bovine seminal vesiculitis [17].

*M. bovisgenitalium* was isolated from the lungs and reproductive tract of dead cows and from aborted fetuses of cattle and buffaloes with arthritis, mastitis

or both [18]. *M. bovis* is known to be a pathological factor associated with bovine necrotic vulvovaginitis, resulting in significant losses to breeders of pedigree livestock [19]. Acute mycoplasmosis causes severe damage to the udder, affecting one to four quarters [8]. Udder hardness and a decrease in milk yield occur. In mastitis, the antibiotic therapy has no treatment effect. In recent years, *M. bovis* has often been isolated from samples of vaginal swabs and respiratory tract of cows with reduced fertility, endometritis, granular vulvitis, or all of these conditions. Also, *M. bovis* is often found in sperm samples and on the mucous membranes of the genital organs in association with *M. bovis* [20].

*M. dispar* is found in the airways of clinically healthy animals. In a study of calves in the Netherlands, *M. dispar* was detected in 92 % of sick and 40 % of healthy animals [21, 22]. In Denmark, *M. dispar* was equally detected in lung samples from calves with signs of fibrinous necrotic purulent bronchopneumonia and embolic pneumonia [23]. Studies of respiratory diseases in dairy calves have shown a possible initiating role of *M. dispar* in the *P. multocida* invasion [24].

*M. dispar* colonizes the mucous membrane epithelium of the respiratory tract, exerting a cytostatic and even cytopathic effect on the cells of the distal bronchi and bronchioles, which impaired tracheobronchial clearance [25]. *M. dispar* causes purple to red consolidations, mainly in the cranioventral areas of the lung [17]. In the UK, *M. dispar* is frequently detected in calves showing respiratory signs. It is believed that the pathogen becomes the main cause of severe pleuropneumonia in adult animals, sometimes with a fatal outcome [26].

**Epizootology.** *M. bovis* was first isolated in the United States in 1961 from cows with severe mastitis [29]. The pathogen has spread to many countries, including Israel (1964), Spain (1967), Australia (1970), France (1974), Great Britain (1974), Czechoslovakia (1975), Germany (1977), Denmark (1981), Switzerland (1983), Morocco (1988), South Korea (1989), Brazil (1989), Republic of Ireland (1994), as well as Northern Ireland (1993) and South Africa (2005) [6].

*M. bovis* is the most economically significant causative agent of bovine mycoplasmosis. Its study was a part of the EU funded DISCONTTOOLS project [30]. In 34 regions of the Russian Federation during 2015 to 2018, *M. bovis* genome was detected in 10.1 %, *M. bovis* genome in 8.6 %, and *M. dispar* genome in 37.15 % of 1186 samples from cows with clinical signs of respiratory, reproductive pathology or both [31].

An outbreak of mastitis due to *M. bovis* was first reported in England in 1960 [18]. Numerous works confirm the spread of this mycoplasma throughout the world. Single cases of *M. bovis* isolation from an aborted horse fetus and boar semen have also been described [32]. *M. bovis* in cattle was detected in the UK, USA, Brazil, Egypt, India, Germany, Austria, Croatia, Denmark, Nigeria, Italy, Japan, Turkey, the Netherlands, Switzerland, South Africa, France, Canada, and Morocco [6, 33].

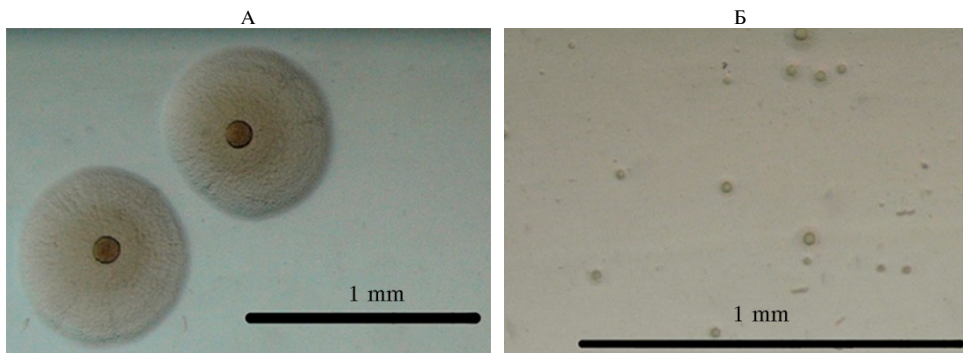
*M. dispar* is one of the pathogens causing respiratory diseases in cattle [34, 35]. *M. dispar* was first isolated in England in 1969 from affected lungs of calves [36]. Then *M. dispar* was found in Denmark, Belgium, Holland, France, Australia, the US, Canada, Korea, and Japan. In Europe, reports of this causative agent came from Great Britain [26]. *M. dispar* infection has been also reported in Brazil [37] and Italy [38].

**Microbiological features.** The identification and detection of mycoplasmas infecting cattle is carried out primarily by microbiological methods. Due to the reduced genome, mycoplasmas cannot synthesize a number of amino acids and, depending on the species, are completely or partially unable to synthesize fatty acids. Mycoplasma receives these nutrients from the host cell.

To culture mycoplasmas, the growing media with bovine heart broth,

serum, yeast extract, peptone, and other additives is necessary with buffering to a final pH of 7.3-7.8 [39]. Examples of nutrient media for mycoplasmas are Edwards nutrient medium (based on bovine heart muscle extract and peptone with horse blood serum and yeast extract), UNIEV medium (tryptic hydrolyzate), Morton's medium (bovine heart extract and bacto-peptone), Hayflick medium (modified Morton's medium added with horse serum and yeast extract).

To suppress growth of other bacteria when isolating the causative agents of mycoplasmosis, the nutrient media are added with antibacterial drugs to which mycoplasmas are insensitive [5]. The growth conditions are 37 °C and 5-10 % CO<sub>2</sub> for 7-10 days. The “fried eggs” appearance is characteristic of most *Mycoplasma* colonies (Fig.), as the central part of a colonies grows into agar while a zone of surface growth is located on the periphery. A number of mycoplasmas have their own characteristic colonies, which makes it possible to differentiate them [39].



Colonies of *Mycoplasma bovis* (3-day culture) (A) and *M. bovisgenitalium* (5-day culture) (B) on solid medium (<https://www.mycoplasma-exp.com/speccultured.html>).

*M. bovis* is not glucose fermenting and does not hydrolyze arginine [40], instead, *M. bovis* uses organic acids such as lactate and pyruvate as energy sources. One of the metabolic products is hydrogen peroxide, an important virulence factor of *M. bovis* [40, 41]. *M. bovisgenitalium* does not hydrolyze arginine and ferment glucose, but has phosphatase activity and reduces tetrazolium salts under anaerobic conditions [42]. *M. bovisgenitalium* also produces hydrogen peroxide [43, 44]. *M. dispar* does not form typical “fried eggs” shaped colonies, especially in the early passages. Growth on nutrient media is slow and requires 7-14 days. *M. dispar* ferments glucose and reduces tetrazolium salts under aerobic and anaerobic conditions, but does not hydrolyze arginine, does not catabolize serum, and does not exhibit phosphatase activity [6, 22, 44].

**Antimicrobial resistance.** Mycoplasmas lack cell walls and thus are resistant to antibiotics which suppress the synthesis of the bacterial cell wall. Mycoplasmas are also resistant to polymyxins, sulfonamides, trimethoprim, nalidixic acid, and rifampicin [45-47]. The antibiotics most commonly used to control mycoplasma infections in cattle are macrolides and tetracyclines. Lincosamides, fluoroquinolones, pleuromutilins, fenicolos, and aminoglycosides may also be active against mycoplasmas [48, 49]. Most strains of mycoplasmas remain sensitive to fluoroquinolones, but pleuromutilins are the most effective [50].

In mycoplasmas, target genes have been identified in which point mutations confer antibiotic resistance, namely, those encoding DNA-gyrase and topoisomerase IV for fluoroquinolones, 23S rRNA for macrolides, lincosamides, pleuromutilins, and amphenicolos, and 16S rRNAs for tetracyclines and aminoglycosides [49, 51, 52].

**Routs of transmission.** The main transmission mechanism is the

aerogenic pathway. Mycoplasmas enter the body through inhalation of contaminated micro-droplets and dust particles. With hematogenous transmission, joint damage is characteristic.

Calves can be infected both horizontally (via aerosols infecting respiratory tract) and vertically from an infected dam. Milk is also among the main sources of infection for calves, especially if the cow has mastitis [53]. With artificial infection of various materials, the survival time of mycoplasmas outside the host is quite long at low temperatures, for example, at 4 °C, the *M. bovis* remains stable in foam for 57 days, in milk for 54 days, on straw for 20 days, on wood and in water for 17 days. The survival period reduced to 1-2 weeks at 20 °C and to 1 week at 37 °C [54].

In a 6-week study of the dairy herd, no evidence of transmission of the pathogen with contaminated sand was found. However, in 12 studied calves, mycoplasmas were detected in samples taken from the upper respiratory tract though the autopsy revealed no signs of pathology [55]. In contrast, another study showed that in a clinical outbreak of *M. bovis*, sand from animal litter without proper treatment may pose a risk of udder infections [56].

In Denmark, studies based on four rounds of milk screening have shown that proximity to farms with confirmed mycoplasmosis increases the risk of introducing the pathogen into a healthy farm. Moving cattle between these farms also poses the risk of infection [57, 58]. Analysis of mycoplasmosis incidence at closely located farms indicates that a rather rapid spread of mycoplasmas if sanitary control measures are not duly observed [59].

**Mycoplasma contamination of sire semen.** *M. bovis* can remain infectious for many years in deep frozen bovine semen. Antimicrobial treatment of semen is ineffective [60].

In in vitro microinjections of *M. bovis* into embryos, the pathogen remained virulent even when thoroughly washed and treated with trypsin and antibiotics (a combination of penicillin, streptomycin, lincomycin and spectinomycin or a combination gentamycin, tylosin, lincomycin and spectinomycin) [61]. In Russia, PCR analysis revealed *M. bovis* DNA in 1.2 %, *M. bovis genitalium* in 43.4 % of 410 sperm specimens from Russian and foreign breeding centers, and the genetic material of *M. californicum* and *Ureaplasma diversum* [62]. The sperm infection by several types of mycoplasmas at once was frequent [62]. In the study of 483 sperm samples from 13 constituent entities of the Russian Federation in 2015-2018, the *M. bovis genitalium* genome was found in 29 %, *M. bovis* in 11.6 % of the specimens [31]. The release of mycoplasmas by infected animals into the environment is not constant, therefore, isolated studies on the detection of a pathogen may be unreliable (63).

The mucous membrane of the upper respiratory tract and mammary glands seem to be the most important site of persistence and shedding of *M. bovis* [64]. Transportation, rearrangement, transfer to the feedlot, and cold stresses cause more aggressive shedding of *M. bovis* from mucous membranes [65]. Chronic asymptomatic infection with intermittent shedding of *M. bovis* is epidemiologically critical [8].

**Incubation period.** Incubation periods of mycoplasma infection largely depend on the age and condition of the animal. In experimental infections, the incubation period is 2-4 days prior to the onset of mastitis and 7 days for pneumonia. In outbreaks of mastitis caused by mycoplasmas, the incubation period was 14 days [66], while an earlier publication reported from 2 to 6 days for similar outbreaks [67]. The incubation period may also depend on the infectious dose, the presence of associated infections, the conditions of the herd, and the

stresses, especially after transportation [68].

**Pathogenesis and its molecular mechanisms.** Mycoplasmas adhere onto host cells to survive as cell surface-associated parasites or further invade into the host cells. Adhesion is an important mechanism of mycoplasma virulence, which is confirmed by the study of avirulent strains unable to adhere [69]. Several adhesive proteins have been identified in *M. bovis*, including the membrane-localized P26 protein [70],  $\alpha$ -enolase, a plasminogen-binding enzyme [71], NADH oxidase [41], and several highly variable membrane proteins (VspA, VspB, VspE, VspF, and VpmaX) [72, 73]. Proteins of the Vsp family spontaneously undergo a random switching between “on” and “off” states of gene expression. The high rate of spontaneous phenotypic Vsp switching is due to frequent rearrangements in the corresponding genes [68].

Such genetic changes affect the virulence of mycoplasmas, biofilm properties [74], susceptibility to phagocytosis and complement-mediated lysis [75], and molecular shielding of antigens [76]. Comparative genomic analysis of different isolates of *M. bovis* revealed many antigenic variations in the surface proteins of the pathogen. The identification and characterization of adhesins contributes to a better understanding of the interactions between *M. bovis* and host cells. It is assumed that fibronectin-binding proteins and a multifunctional glycoprotein of the extracellular matrix can stimulate adhesion of mycoplasmas to host cells [77]. Several of these proteins have been identified in mycoplasmas. For example, the *M. bovis* TrmFO protein plays an important role in the adhesion of a pathogen to cells, and also performs the function of tRNA methyltransferase [77]. Another distinctive feature of mycoplasmas is their ability to penetrate and multiply in various types of cattle mononuclear cells in the peripheral blood and in erythrocytes, which allows the pathogen to evade host immune system and antimicrobial drugs, and also contributes to rapid spreading in the body of an infected animal [78]. *M. bovis* antigens were visualized in hepatocytes and epithelial cells [79], and invasion accompanied by intracellular replication was demonstrated in epithelial cells of the respiratory tract of cattle embryos [80].

The ability of many mycoplasma species to form biofilms is noteworthy [74]. Despite the fact that the biofilms were mainly demonstrated in vitro [74, 81, 82], some researchers report that the mycoplasma biofilms can affect the course of the disease or the pathogenicity of microorganisms in cattle [83, 84]. Nevertheless, the genes encoding the ability to form biofilms characteristic of other bacterial species were not found in mycoplasmas.

In *M. mycoides* subsp. *mycoides*, key proteins associated with biofilm formation have been identified, such as elongation factor Tu, glucose-specific transporter IIB of the PTS system, phosphoenolpyruvate protein phosphotransferase, fructose bisphosphate aldolase II, and pyruvate dehydrogenase [85]. A comparative study of different isolates of *M. bovis* revealed correlation between biofilm formation and the corresponding Vsp expression profile. The ability to develop biofilms can explain the *M. bovis* survival in the litter [56, 86].

To date, only one study has reported putative role of specific genes in pathogenicity of *M. bovis*. When comparing the reduced pathogenicity in the 115th, 150th and 180th passages of the *M. bovis* isolate as compared to its parent wild-type isolate *M. bovis* HB0801, 11 genes were identified that affect the attenuation process. Of these, 10 are associated with metabolism and one encodes a variable surface protein [87]. However, in vivo, no gene has been proven to influence the virulence profile of *M. bovis* [68].

**Laboratory diagnostics.** Lab tests are critical for clinical diagnosis, since bovine mycoplasma-induced clinical signs are not pathognomonic. The

etiology of *M. bovis* is often overlooked until other pathogens have been ruled out or animals stop responding to antibiotic therapy [68, 88]. Culture, molecular, and serological methods are used to identify *M. bovis* and other types of cattle mycoplasmas in milk, articular fluid, bronchoalveolar lavage fluid, smears from mucous membranes or sera [89]. Identification of mycoplasma isolates to species is extremely important, since species such as *M. bovis genitalium*, *M. bovis*, and *M. dispar* are classified as primary pathogens while others are deemed part of a resident microbiome that does not play a significant role in the development of diseases [6].

Culture, as a conventional microbiology method, is the standard for the detection of mycoplasmas, but it takes a long time [18] and requires enriched media and antibiotics [68]. There is also a risk of false positives due to the similarity of *M. bovis* colonies to colonies of other bacteria of the same class. Also, this method is applicable for viable microorganisms [20]. The sensitivity and specificity of the microbiology testing have not been determined for the majority of bacteria involved in the development of respiratory diseases in cattle [88]. A 70.7 % sensitivity and 93.9 % specificity have been shown for growing *M. bovis* on a solid medium with Tween 80 [90].

Serological methods are used to assess the effectiveness of vaccination, to determine the immune status of the herd and the spread of infection on a wider scale [91]. The enzyme-linked immunosorbent assay (ELISA) is a tool only for indirect confirmation of infection, since it detects antibodies to *M. bovis* but not the pathogen itself [20]. ELISA is recommended in combination with other methods to minimize false positives due to the presence of antibodies to *M. bovis* in healthy animals [20, 88].

Over the past two decades, the polymerase chain reaction (PCR) has been recognized as the main test for diagnosing mycoplasmosis [88, 92]. However, PCR requires specific oligonucleotide primers designed based on sequencing genomes of the pathogens [88]. PCR analysis, as a rule, is expensive, but pooled samples allow a group of animals to be tested [93-95].

In general, molecular methods can be divided into those that allow the detection of a specific type of mycoplasma (using classical PCR or real-time PCR) [96, 97] or mycoplasmas without determining their species. The methods enabling differentiation of mycoplasma species are multiplex PCR [98, 99], PCR with denaturing gradient gel electrophoresis [100], DNA microarrays, and multilocus sequence typing [38, 101].

In the last decade, the matrix assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS) which enables identification of bacteria by their unique protein profiles is recognized a revolutionary milestone in diagnostics. The method is mainly used for identification of cultured bacteria and mycoplasmas [91, 102], but it is less effective in polymicrobial samples and in samples with mixed infection.

Recently, next generation sequencing (NGS) technique has been widely used to study the bacterial genome. NGS data are used for clinical diagnostics, in the study of outbreaks of diseases, and in the control of the microbial resistance to antimicrobial drugs [104]. To date, complete genome sequences are available for five isolates of *M. bovis* [105-107], two isolates of *M. californicum* [108, 109], and for *M. arginini* [110], *M. bovis genitalium* [111], *M. canadense* [112], *M. bovoculi* [113], *M. leachii* [114], one isolate each.

In addition to the described methods, latex agglutination test and immunochromatographic analysis are under development [68]. A number of studies have recently reported application of a loop-mediated isothermal amplification assay for the detection of *M. bovis* [115, 116].

Thus, *Mycoplasma bovis*, *M. bovis genitalium* and *M. dispar* are the most important members of the *Mollicutes* class which affect cattle. They cause many diseases, the most significant of which are mastitis, pneumonia, reproductive disorders and arthritis in calves and adult animals. These diseases can be chronic, reducing the resistance of cattle to other viral and bacterial pathogens. Mortality and morbidity can reach 10 and 35 %, respectively. Mycoplasma infections has a negative economic impact on animal farming. The herd mostly becomes infected from animals with a subclinical course of the disease. However, there are many other routes to transmit mycoplasmas. The chronic character of mycoplasma diseases and subclinical forms hamper the identification of the infection. The resistance of mycoplasmas to a large number of antimicrobial drugs complicates antibiotic therapy so much that for some diseases, such as mastitis, it is now recommended to slaughter all affected animals. The complexity of the treatment of mycoplasmosis determines the relevance of prevention measures. Unfortunately, the development of a mycoplasma vaccine is a difficult task, and so far, such vaccines cannot be considered as a tool for the control of mycoplasma infections in cattle. Epidemiological studies have identified the main factors of biosecurity risk for the spread of mycoplasmosis, e.g., the introduction of new cattle and poor milking hygiene. Timely and accurate diagnosis contributes to the prevention of the disease. However, additional research and effective control programs for pathogenic mycoplasmas are required to fully understand the mechanism of mycoplasma spread.

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