ОШ МАМЛЕКЕТТИК УНИВЕРСИТЕТИНИН ЖАРЧЫСЫ. АЙЫЛ ЧАРБА: АГРОНОМИЯ, ВЕТЕРИНАРИЯ ЖАНА ЗООТЕХНИЯ

ВЕСТНИК ОШСКОГО ГОСУДАРСТВЕННОГО УНИВЕРСИТЕТА. СЕЛЬСКОЕ ХОЗЯЙСТВО: АГРОНОМИЯ, ВЕТЕРИНАРИЯ И ЗООТЕХНИЯ

JOURNAL OF OSH STATE UNIVERSITY. AGRICULTURE: AGRONOMY, VETERINARY AND ZOOTECHNICS

e-ISSN: 1694-8696
№2(11)/2025, 30-43

ВЕТЕРИНАРИЯ

УДК: 619.616.98:579.852.13.
DOI: 10.52754/16948696_2025_2(11)_3

STUDY OF THE EPIZOOTOLOGY OF SHEEP BRUCELLOSIS IN SOME REGIONS OF UZBEKISTAN AND THE RESULTS OF DIAGNOSTIC STUDIES

ИЗУЧЕНИЕ ЭПИЗООТОЛОГИИ БРАДЗОТА ОВЕЦ В НЕКОТОРЫХ РЕГИОНАХ УЗБЕКИСТАНА И РЕЗУЛЬТАТЫ ДИАГНОСТИЧЕСКИХ ИССЛЕДОВАНИЙ

ӨЗБЕКСТАНДЫН АЙРЫМ АЙМАКТАРЫНДА КОЙЛОРДУН БРАДЗОТУНУН ЭПИЗООТОЛОГИЯСЫН ИЗИЛДӨӨ ЖАНА ДИАГНОСТИКАЛЫК ИЗИЛДӨӨЛӨРДҮН ЖЫЙЫНТЫКТАРЫ

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Abstract

The development of sheep breeding as one of the leading areas of animal husbandry in Uzbekistan requires a systematic approach to the prevention of particularly dangerous infectious diseases, including sheep botulism caused by Clostridium oedematiens. The relevance of this study is due to the need for timely detection and prevention of outbreaks of this infection, which leads to significant economic losses and the death of productive animals. The aim of the study was to establish the epizootic situation regarding sheep botulism in the Kashkadarya, Samarkand and Jizzakh regions, as well as to conduct a comprehensive diagnosis of the disease, followed by the isolation and study of the biological properties of the pathogen. To achieve this goal, visits were organised to disadvantaged private and karakul sheep farms, where pathological material was collected from dead animals. Diagnostics included bacteriological studies using Kitt-Tarozzi medium, Gram staining, microscopy, and biological testing on laboratory animals. It was established that brucellosis in these regions was mainly recorded during the cold season – autumn, winter and early spring. The disease mainly affects sedentary and well-fed sheep, especially when grazing in conditions of high humidity. The isolated strains of Cl. septicum demonstrated characteristic growth with clouding of the medium and the formation of gas bubbles. Laboratory animals infected with the isolated culture showed typical clinical and pathological changes, confirming the high pathogenicity of the pathogen. The diagnosis was based on a combination of epizootological data, laboratory tests and bioassays. The results of the study are of practical importance for determining vaccination schedules, planning anti-epizootic measures and preventing economic losses in sheep breeding

Keywords: pathogen; Cl. septicum; bacillus; spore; anaerobe; Kitt-Tarozzi; sheep breeding

Изучение эпизоотологии брадзота овец в некоторых регионах Узбекистана и результаты диагностических исследований

Аннотация

Развитие овцеводства как одного из ведущих направлений животноводства Узбекистана требует системного подхода к профилактике особо опасных инфекционных заболеваний, в числе которых брадзот овец, вызываемый Clostridium oedematiens. Актуальность настоящего исследования обусловлена необходимостью своевременного выявления и предупреждения вспышек данной инфекции, приводящей к значительным экономическим потерям и падежу продуктивных животных. Целью работы являлось установление эпизоотической обстановки по брадзоту овец в Кашкадарьинской, Самаркандской и Джизакской областях, а также проведение комплексной диагностики заболевания с последующим выделением и изучением биологических свойств возбудителя. Для достижения цели были организованы выезды в неблагополучные частные и каракулеводческие хозяйства, где от павших животных отбирался патологический материал. Диагностика включала бактериологические исследования с использованием среды Китта-Тароцци, окраску мазков по Граму, микроскопию, а также биологическую пробу на лабораторных животных. Установлено, что брадзот в указанных регионах регистрировался преимущественно в холодное время года – осенью, зимой и ранней весной. Заболеванию подвержены,

Өзбекстандын айрым аймактарында койлордун брадзотунун эпизоотологиясын изилдөө жана диагностикалык изилдөөлөрдүн жыйынтыктары

Аннотация

Өзбекстандагы мал чарбачылыгынын негизги багыттарынын бири болгон кой чарбачылыгынын өнүгүшү өзгөчө коркунучтуу инфекциялык оорулардын алдын алуу боюнча системалуу мамиленин зарылдыгын туудурат. Алардын катарында Clostridium oedematiens бактериясынын түрткү болгон койлордун брадзоту бар. Учурдагы изилдөөнүн актуалдуулугу – аталган инфекциянын кырдаалдарын өз убагында аныктоо жана алдын алуу зарылдыгында, анткени бул оорулуу экономикалык чыгымдарды көбөйтүп, продуктивдүү малдардын өлүмүнө алып келет. Изилдөөнүн максаты – Кашкадария, Самарканд жана Джизак облустарында койлордун брадзотунун эпизоотологиялык абалын аныктоо, ооруну комплекстүү диагностикалоо, оорунун козгогучун бөлүп алуу жана анын биологиялык касиеттерин изилдөө болду. Максатка жетүү үчүн көйгөйлүү жеке жана каракул чарбаларына барышып, каза болгон малдардан патологикалык материал алынды.

Диагностика Китт-Тароцци чөйрөсүндө бактериологиялык изилдөөлөрдү, Грамга боёону, микроскопияны жана лабораториялык жаныбарларда биологиялык сыноону камтыды. Аймактарда брадзот негизинен суук мезгил – күз, кыш жана эрте жаз айларында катталган. Ооруруга негизинен аз кыймылдуу жана жакшы салмак алган койлор сезгентилген, айрыкча жогорку нымдуулуктагы

главным образом, малоподвижные и хорошо упитанные овцы, особенно при пастьбе в условиях повышенной влажности. Выделенные штаммы Cl. septicum продемонстрировали характерный рост с помутнением среды и образованием газовых пузырьков. У лабораторных животных, инфицированных выделенной культурой, наблюдались типичные клинические и патологоанатомические изменения, подтверждающие высокую патогенность возбудителя. Диагноз устанавливался на основе совокупности эпизоотологических данных, лабораторных тестов и биопробы. Результаты исследования имеют практическое значение для обоснования сроков вакцинации, планирования противоэпизоотических мероприятий и предотвращения экономических потерь в овцеводстве

Ключевые слова: возбудитель; Cl. septicum; бацилла; спора; анаэроб; Китт-Тарощци; овцеводческое хозяйство

жайыттарда. Бөлүнүп алынган Clostridium septicum штаммдары өзгөчө өсүү менен, чөйрөнүн булануусу жана газ көбүкчөлөрүнүн пайда болушу менен мүнөздөлдү. Лабораториялык жаныбарларда бөлүнгөн культура менен жугузулган учурда типтүү клиникалык жана патологоанатомиялык өзгөрүүлөр байкалган, бул козгогучтун жогорку патогендигин тастыктады. Диагноз эпизоотологиялык маалыматтардын, лабораториялык тесттердин жана биологиялык сыноонун комплексинин негизинде коюлду. Изилдөөнүн жыйынтыктары эмдөөнүн мөөнөттөрүн белгилөөдө, эпизоотияга каршы чараларды пландаштырууда жана кой чарбачылыгындагы экономикалык жоготуулардын алдын алууда практикалык мааниге ээ

Ачкыч сөздөр: козгогуч; Cl. septicum; бацилла; спора; анаэроб; Китт-Тарощци; кой чарбасы

Introduction

Sheep brucellosis remains one of the serious factors limiting the development of karakul sheep farms and private households in the Republic of Uzbekistan. In this regard, the development of resource-efficient and import-independent preventive measures to combat infectious animal diseases is particularly relevant. Since 2018, large-scale measures have been implemented in the republic to prevent and control anaerobic infections affecting large and small ruminants. Among the key tasks in the field of animal husbandry, the priorities remain increasing livestock numbers, improving productivity, ensuring full reproduction, proper animal care and the prevention of dangerous diseases.

In order to ensure the sustainable development of karakul sheep breeding, increase the number of karakul sheep, expand production and processing, improve breeding work and social support for industry specialists, a set of regulatory and legal acts regulating state support for the sector was adopted. In particular, Decree of the President of the Republic of Uzbekistan No. DP-60 (2022), Resolution of the President of the Republic of Uzbekistan No. RP-3603 (2018), and Decree of the President of the Republic of Uzbekistan No. PP-224 (2022) are aimed at addressing the above-mentioned tasks.

However, despite the creation of additional opportunities and conditions, the problem of developing and increasing the productivity of sheep breeding remains widespread. The study by H. Esmaeili et al. (2025) examines the clinical and laboratory aspects of malignant oedema in sheep caused by Clostridium septicum, a pathogenic anaerobe similar in characteristics to the causative agent of brucellosis. Based on the study of clinical cases in imported sheep breeds (Il-de-France and Roman) in three farms in Iran, the authors conducted a comprehensive diagnosis, including bacteriological studies, Gram staining and PCR identification of the pathogen. The high pathogenicity of C. septicum and the need for early diagnosis of the disease were established, as its course is rapid and leads to death. It was noted that despite the availability of vaccines, the disease remains insufficiently studied and covered in veterinary practice.

A. Mussayeva et al. (2024) analysed outbreaks of brucellosis and anaerobic enterotoxemia in sheep in various regions of Kazakhstan in 2021-2022. Pathological material was examined using Kitt-Tarozzi medium, glucose-blood agar, microscopy and bioassays on guinea pigs. The cultural and morphological properties of Clostridium perfringens (types A, B, SVT, D), Cl. septicum and Cl. oedematiens were characterised. Over the decade, 15 cases of botulism and 44 cases of enterotoxemia were identified, with the majority occurring in the Zhambyl region. The authors attribute the low incidence to systematic vaccination, emphasising the importance of prevention and its potential as a model for other countries.

The article by R.M. Uraqova & H.S. Salimov (2021) presents experimental data on the determination of 50% and 100% lethal doses (LD_{50} and LD_{100}) of the causative agent of brucellosis – Clostridium oedematiens in guinea pigs. The bacteria were isolated from pathological material from sheep that had died from brucellosis and cultured in Kitt-Tarozzi medium. The authors emphasised that the most important predisposing factors for the development of the disease are winter and early spring grazing of sheep in high humidity, helminth infection, and deficiency of proteins, vitamins, and minerals, which leads to a decrease in the overall resistance of the organism. It has been established that Cl. oedematiens is a motile anaerobic microorganism capable of sporulation in the body of a dead animal. Infection occurs through feed, water, soil and manure. The

results of laboratory determination of lethal doses are important for understanding the pathogenesis and toxicity of isolated local strains, as well as for the development of effective preventive and diagnostic measures against brucellosis in sheep.

P. Thomas et al. (2021) presented the first complete genomic sequence of the C. septicum DSM 7534T strain and performed a comparative analysis of five genomes of this species. The data revealed a stable phylogenetic position of C. septicum, significant variability in prophages, CRISPR elements, and restriction-modification systems, as well as the presence of an extensive pool of virulence genes, including sialidase, collagenase, haemolysin, and leukocidin homologue genes. These results shed light for the first time on the genetic plasticity and pathogenicity of C. septicum, expanding understanding of its role as a dangerous zoonotic pathogen.

To develop effective measures to combat brucellosis, it is necessary to study its epizootiology and improve methods for diagnosing and preventing the disease. Of particular importance in this regard was the isolation of local strains of the pathogen from pathological material taken from sheep in farms affected by brucellosis and the study of their cultural, morphological and biological characteristics. The aim of the study was to isolate the pathogen from pathological material from sick and dead animals, to study its cultural-morphological, tinctorial, biochemical and biological properties in order to substantiate the diagnosis and develop preventive measures.

Materials and methods

The study was conducted in 12 sheep farms located in the Kashkadarya, Samarkand and Jizzakh regions of the Republic of Uzbekistan. The total number of sheep in the surveyed farms was about 8,000, which corresponds to the number of sheep in focal flocks with an increased incidence of brucellosis. At the same time, the total number of sheep in the three regions is about 50-70 thousand. For bacteriological studies, pathological material (75 samples) was collected from dead animals (Fig. 1): pieces of liver, heart, kidneys, tubular bones and muscle tissue from the affected areas (Fig. 2). Samples were collected in accordance with the rules of asepsis and antisepsis.

Figure 1. Sheep dead from brucellosis

Source: photos taken by the authors

Figure 2. Damaged internal organs

Source: photos taken by the authors

In the immunology and biotechnology laboratory of the Veterinary Research Institute (Tashkent, Uzbekistan), pathological material was subjected to bacteriological examination. For microscopic analysis, smears were prepared on microscope slides from the organs under investigation. The smears were air-dried and fixed over the flame of an alcohol lamp, which ensured their sterility and prevented bacterial growth during subsequent staining and microscopy. After fixation, the smears were stained according to Gram, which allowed microorganisms to be differentiated into Gram-positive and Gram-negative. The staining technique included the sequential application of carbolic acid solution of gentian violet (1-2 minutes), treatment with Lugol's solution (1 minute), decolorisation with alcohol (about 30 seconds), additional staining with an alcohol-water solution of fuchsin (1-2 minutes), after which the preparation was washed with water and dried (Eleusizova, 2018). To isolate bacteria, the surface of the organs was burned with a heated scalpel, then tissue fluid was collected from the tissues using a Pasteur pipette and seeded into test tubes with Kitt-Tarozzi medium (Litusov, 2015). The test tubes were incubated in a thermostat at 37-38 °C for the growth of microorganisms.

To confirm the pathogen and make a diagnosis, biological tests were carried out on laboratory animals (guinea pigs), which were infected with a daily culture of the isolated pathogen, Clostridium septicum. Clinical signs, pathological changes and the results of bacteriological studies were observed. Epizootological data were collected during business trips to the farms under study, together with local veterinary specialists, where information on the spread of the disease, the conditions of keeping and the nature of the affected animals was recorded.

To prevent and control brucellosis, sheep were vaccinated twice 35-40 days before being taken to pasture, with an interval of 12-14 days. The vaccine doses were: 2 and 3 ml for adult sheep, 1 and 1.5 ml for lambs up to 6 months old, respectively, administered intramuscularly into the inner

hairless surface of the hind limb. Immunity developed 10-12 days after the second dose (Dunlop & Malbert, 2004).

When the disease was detected on a farm, restrictions were imposed: a ban on the import and export of animals, and on the movement or transport of healthy unvaccinated sheep. Sanitary measures were carried out – destruction of carcasses, manure and feed residues by burning, disinfection of the territory with solutions of chlorinated lime (3%), formaldehyde (5%) and caustic soda (5%) with re-treatment after one hour.

During the research, all applicable ethical standards and requirements governing the use of animals for scientific purposes were observed. All manipulations with animals were carried out in strict accordance with the regulatory documents of the Republic of Uzbekistan and international standards for the treatment of laboratory and farm animals (International Standards for the Care..., 2019). Biological tests on guinea pigs were carried out using the minimum number of animals necessary to achieve reliable results. Before the start of the research, a positive review (permission) was obtained from the ethics committee of the Research Institute of Veterinary Medicine.

Results

The disease was recorded in various pastures, pens and subsidiary farms, mainly during sudden changes in weather conditions and violations of sanitary and hygienic standards for animal husbandry. Among the sheep examined in individual flocks, a high incidence of brucellosis was detected – in some cases it reached about 30%, while the mortality rate among those affected remained extremely high – 90-100%. The incidence rate among the total sheep population in the regions studied was estimated at approximately 1-1.5%. The highest number of cases was recorded in the Shakhrisabz district of the Kashkadarya region, the Payaryk district of the Samarkand region and the Gallyaaral district of the Jizzakh region, as shown in Figure 3.

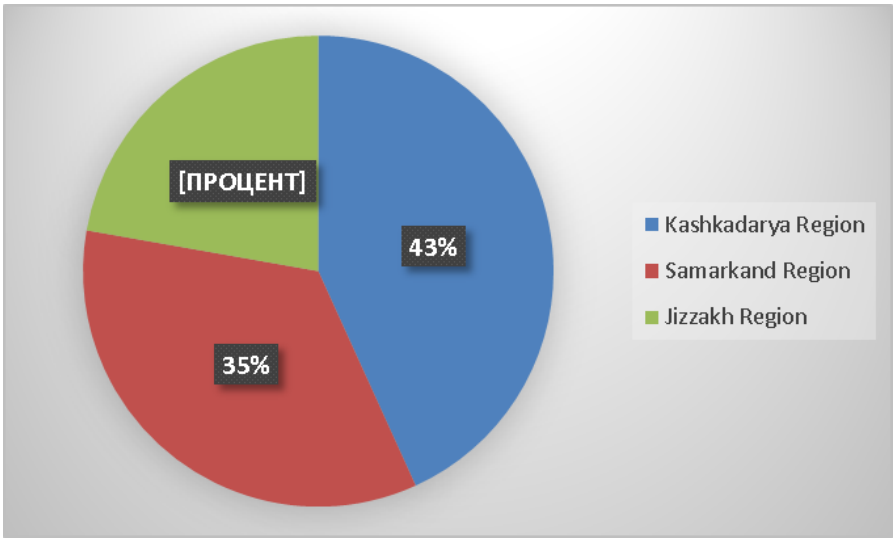

Figure 3. Prevalence of brucellosis by region (number of sheep affected)

Source: compiled by the authors

Based on the data presented in Figure 3, the highest epizootic intensity was observed in the Shakhrisabz district of the Kashkadarya region, where approximately 350 sheep were reported to be infected. A significant number of cases were also detected in the Payaryk district of the Samarkand region (approximately 280 animals) and the Gallyaaral district of the Jizzakh region (approximately 180 animals). This distribution indicates a probable dependence of the spread of brucellosis on

environmental and economic-organisational factors. The disease was particularly intense in areas with a high density of sheep farms and inadequate sanitary control over housing conditions. Increased susceptibility of animals was observed during spring and summer fluctuations in temperature and humidity, which apparently contributed to the activation of the pathogen and the occurrence of acute outbreaks.

The studies established that sedentary and well-nourished sheep were highly susceptible to the disease, becoming infected mainly when grazing on pastures, whereas lambs contracted the infection in pens or household conditions. Despite the absence of a pronounced seasonal dependence, brucellosis in sheep is more often recorded between October and March. The main predisposing factors are winter and early spring grazing in pastures with high air humidity – the presence of dew, frost and precipitation, which creates favourable conditions for the contamination of feed and pastures with the pathogen.

An additional risk factor is a decrease in the overall resistance of the body caused by helminthiasis and disturbances in protein, vitamin and mineral metabolism. Immunodeficiency states contribute to the penetration of Clostridium septicum into the digestive organs and the accelerated development of the pathological process. Thus, temperature and humidity conditions, combined with the physiological weakening of sheep, create a favourable background for the development of brucellosis and high mortality among affected animals.

To confirm the diagnosis and clarify the aetiology of the disease, laboratory tests were carried out on pathological material taken from dead and clinically ill animals in outbreak areas. The material under investigation was sent to the Laboratory of Immunology and Biotechnology of the Research Institute of Veterinary Medicine, where bacterioscopic and bacteriological analyses were performed. Particular attention was paid to the isolation of a pure culture of the pathogen and the determination of its morphological and cultural properties. Figure 4 shows the growth of the causative agent of brucellosis in anaerobic conditions, accompanied by clouding of the nutrient medium and the formation of gas bubbles under petroleum jelly oil.

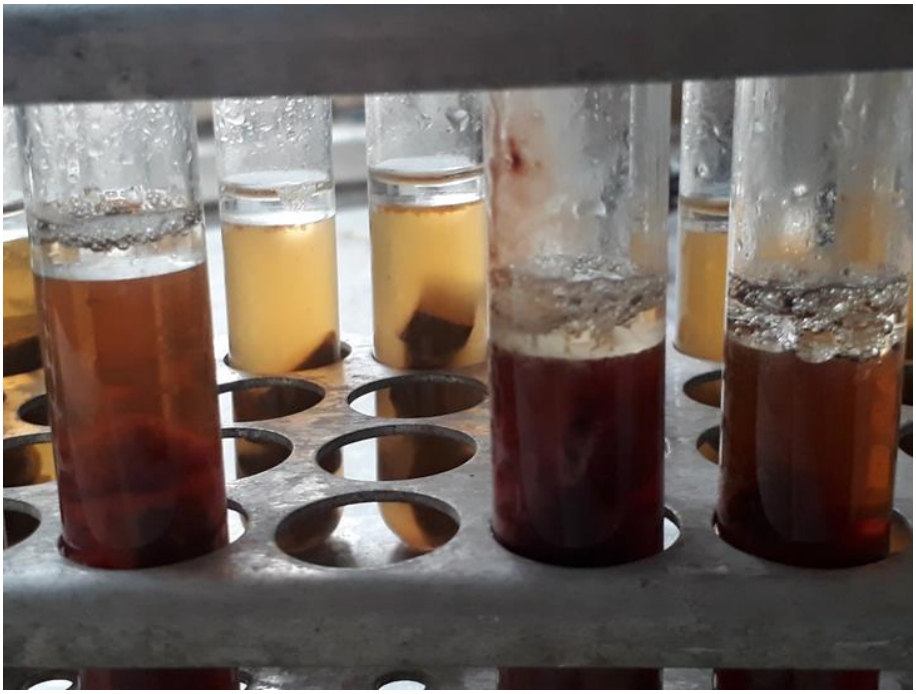

Figure 4. Growth of the causative agent of blight with clouding of the culture medium and formation of air bubbles under petroleum jelly oil

Source: compiled by the authors

The figure shows test tubes with meat-peptone broth inoculated with pathological material taken from dead sheep in areas suspected of having brucellosis. Incubation was carried out under anaerobic conditions at a temperature of 37°C for 24-48 hours. Characteristic signs of obligate anaerobic growth are visually observed: clouding of the medium, flaky sediment and the formation of gas bubbles under a layer of petroleum jelly oil, indicating active metabolic activity of the pathogen. The change in the colour of the medium from light yellow to dark brown may be due to the formation of metabolites and protein breakdown products accompanying the vital activity of clostridia and confirms the reproduction of the pathogen and its interaction with the components of the culture medium. However, the colour of the medium is not a specific diagnostic feature. Thus, the results of bacteriological culture confirm the presence of Clostridium septicum in the test material and indicate a typical course of blackleg in sheep, accompanied by massive intoxication, gas formation and high mortality. A microscopic image of Gram-positive bacteria of the causative agent of brucellosis is shown in Figure 5.

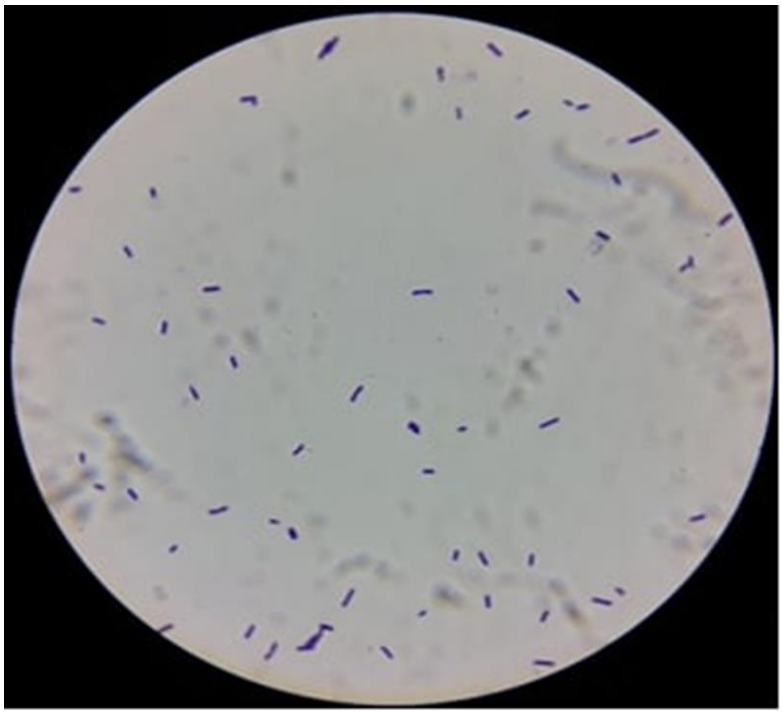

Figure 5. Microscopic image of the causative agent of brucellosis, stained according to Gram

Source: compiled by the authors

Microscopic examination of stained preparations prepared from pathological material revealed characteristic morphological features of the pathogen. Figure 5 shows Gram-positive rod-shaped bacteria, located mainly singly or in pairs, with clear contours and a characteristic purple colour, indicating the resistance of the cell wall to the action of alcohol during Gram staining. The bacteria ranged in size from 10 to 30 μm in length, with uneven thickness and a moderately pronounced capsule structure (Ferreira Alves et al., 2021). The microscopic data obtained indicate that the microorganisms belong to the genus Clostridium, and the combination of morphological features confirms the identification as Clostridium septicum, the causative agent of malignant oedema and brucellosis in sheep.

For final verification of the diagnosis, a biological test was performed: laboratory guinea pigs were injected with a daily culture isolated from pathological material obtained from dead sheep.

Characteristic clinical manifestations, pathological changes, and the results of subsequent bacteriological examination of infected animals confirmed the presence of Cl. septicum and served as the basis for the final diagnosis. After laboratory confirmation of the diagnosis, measures were promptly taken to treat and prevent the disease in the focus of infection. Sheep are vaccinated 35-40 days before the flock is put out to pasture. Immunisation is carried out twice at intervals of 12-14 days using a specific vaccine against brucellosis. Recommended dosages: adult animals – 2 ml for the first injection and 3 ml for the second; lambs up to 6 months of age – 1 ml and 1.5 ml, respectively. The vaccine is administered intramuscularly into the inner hairless surface of the hind limb. Immunity develops 10-12 days after the second injection.

If a case of brucellosis is confirmed on a farm, quarantine restrictions are imposed: the import and export of animals, as well as the movement and transport of unvaccinated sheep on the farm, are prohibited. All dead animals, feed residues and manure must be destroyed by burning. The territory is subjected to sanitary cleaning, and then disinfected twice, at intervals of one hour, with a 3% solution of bleach, a 5% solution of formaldehyde or a 5% solution of caustic soda.

Thus, comprehensive epizootological, laboratory and pathological studies have reliably established that Clostridium septicum is the causative agent of the reported cases of disease in sheep in a number of regions of Uzbekistan. Microscopic, bacteriological and biological methods, used in combination, ensured a high degree of diagnostic accuracy. The established seasonal patterns, clinical and anatomical picture and laboratory test results made it possible to formulate scientifically sound recommendations for the prevention, localisation and elimination of infection foci.

Discussion

The presence of Clostridium septicum in abomasal samples, despite its low detection rate, was confirmed by data from H. Kalender et al. (2023), who identified C. septicum among a number of clostridia in Turkey, albeit with a low prevalence rate, which was associated with mass vaccination of cows. In the conditions of the study in Uzbekistan, clinical foci were characterised by a high density of infection and the absence of large-scale prevention, which, apparently, led to a higher proportion of infected animals. Thus, the results showed that vaccination significantly reduced the frequency of C. septicum detection in the abomasum, which correlated with observations on the need for preventive measures.

The textbook by A.A. Shevchenko et al. (2013) was devoted to the study of anaerobic enterotoxemia in sheep and goats caused by Clostridium perfringens types C and D. The authors summarised a large amount of data on the pathogenesis, prevalence and methods of combating the disease. The work examined the historical development of the study of the disease, its economic damage, the resistance of the pathogen, as well as clinical and pathological signs. Particular attention was paid to diagnostic methods, including bacteriological studies, differential diagnosis, and laboratory confirmation. Based on the data obtained, recommendations were made for treatment, vaccination, and sanitary and preventive measures in affected farms.

A study by S. Moustafa et al. (2022) in Egypt showed that typical toxin-producing strains of C. perfringens were more prevalent in haemorrhagic abomasitis, while C. septicum was significantly less common. These data allowed a clear distinction to be made between brucellosis and abomasal enteritis, emphasising that the toxigenicity of C. septicum was mainly manifested in

other clinical forms, such as malignant oedema and brucellosis, which was confirmed by observations in Uzbekistan. The results obtained complemented the understanding of the pathogenetic differences between clostridia and clarified the specificity of C. septicum in various forms of the disease.

W.-X. Tan et al. (2025) reported a high prevalence of C. perfringens among sheep in the Asian region (~38.8%) and demonstrated the effectiveness of vaccination in reducing morbidity. This was consistent with the idea of the key role of preventive measures in enterotoxemia. Despite the difference in the aetiological agent – C. perfringens versus C. septicum – both pathogens were activated under the influence of stress factors associated with feeding and climatic changes. Consequently, the conclusion about seasonal outbreaks caused by humidity and temperature was confirmed in a broader regional context.

The review by Y.K. Prabhakar et al. (2025) presented modern molecular biological aspects of the epidemiology, pathogenesis, immunobiology, genomics, and proteomics of brucellosis pathogens in cattle. The authors described in detail the mechanism of severe inflammatory changes in the reproductive organs of animals, confirmed by the detection of vasculitis, fibrosis, and necrosis. Of particular importance was the consideration of the immune mechanisms by which the pathogen evades the host response, as well as the use of proteomics to search for biomarkers and potential therapeutic targets. Unlike the present study, which focused on sheep brucellosis, this review demonstrated a more comprehensive molecular approach to characterising the infection, including genomic sequencing and drug resistance analysis.

A detailed analysis of the clinical and pathological course of brucellosis in sheep was presented by J.F. Prescott et al. (2017), who considered the disease to be a form of clostridial abomasitis caused by Clostridium septicum. Characteristic macroscopic and microscopic changes were described, including oedema, gas formation in the abomasal wall, pronounced necrosis and vascular thrombosis, which fully coincided with the results recorded during outbreaks in Uzbekistan. The role of cold climate and damage to the mucous membrane as factors contributing to the development of infection was also emphasised. Particular attention was paid to rapid mortality, diagnostic difficulties in post-mortem isolation of the pathogen, and the limited effectiveness of antibiotic therapy, which coincided with conclusions about the need for vaccination and strict sanitary control. Unlike the present study, the presented work focused on experimental data on modelling the pathogenesis and international spread of the disease.

A study by S. Ting et al. (2025) conducted in Timor-Leste revealed low levels of awareness among farmers about brucellosis, but high willingness to vaccinate and use personal protective equipment. The authors pointed out that domestic and traditional practices, such as grazing on common pastures and leaving the placenta in the field, significantly increased the risk of spreading the infection to both animals and humans. These data correlated with the findings of the present study on the role of sanitary violations and pasture conditions in the spread of brucellosis. Despite the difference in aetiology – Brucella spp. in Timor and Clostridium septicum in this study – both works emphasised the key role of animal hygiene and epizootiological literacy in disease prevention.

R.M. Ortiz Flores et al. (2024) demonstrated that partially purified protein fractions secreted by Clostridium septicum induced macrophage death through the activation of autophagy and apoptosis. It was found that proteins with a molecular weight ≥100 kDa caused pronounced

inflammation, morphological changes in cells, and increased levels of IL-10 and TNF-α, indicating an attempt by the pathogen to evade the immune response at the site of infection. The data obtained confirmed the pathogenic potential of C. septicum as a highly virulent pathogen, which was fully consistent with the high mortality and rapid progression of brachitis in sheep observed in the present study. In contrast to the clinical focus of the current work, the article by R.M. Ortiz Flores et al. provided a molecular explanation for the pathogen's ability to cause massive tissue damage and immune dysfunction, expanding the understanding of the mechanisms of C. septicum virulence.

Scientists Z. Kirimbayeva et al. (2023) conducted epizootic monitoring of bacterial infections in animals in the southern regions of Kazakhstan, with a particular focus on clostridiosis, including emphysematous carbuncle. The study noted that these diseases mainly occurred during the grazing period, which correlated with the findings of the present study on outbreaks of brucellosis in sheep under similar conditions. In both cases, the role of weather factors, pasture structure and stocking density in the spread of infection was emphasised. Despite the low level of positive soil samples for clostridia (less than 1%), the authors pointed to the need for further research into the influence of the environment on morbidity. The difference was in the broader coverage of pathogens (including Listeria spp.) and the use of government monitoring data, whereas the present study relied on laboratory and field epizootiological methods.

Thus, a summary of the results of various independent studies conducted in Turkey, Kazakhstan, Brazil, and Egypt confirmed the key findings of this study: climatic conditions (humidity, temperature), poor sanitation, immunodeficiency, and lack of vaccination were the main predisposing factors for the development of brucellosis in sheep. The similarity of the data obtained with the results of other studies strengthened the evidence base, confirming the key factors in the development of the disease. At the same time, geographical and pathogenetic differences only emphasised the regional specificity of the pathology and the need to develop adapted prevention strategies.

Conclusions

The study found that brucellosis was recorded among sheep mainly in farms with foci of infection in certain areas of the Kashkadarya, Samarkand and Jizzakh regions of the Republic of Uzbekistan. In some of the flocks examined, the proportion of infected animals reached 30%, indicating the high intensity and focal nature of the disease. The mortality rate among infected sheep was 90-100%, reflecting the severity and acute course of the disease.

The overall prevalence of brucellosis in the regional sheep population, taking into account all the farms surveyed, is estimated at 1-1.5%. These figures are based on epizootological monitoring and a comprehensive analysis of clinical and laboratory studies. The difference between the high incidence in foci and the relatively low prevalence overall is explained by the localisation of infection foci and the effectiveness of preventive measures in most farms.

The highest number of cases was registered in areas with a high density of sheep farms and insufficient sanitary and hygienic control, which indicates the significant role of economic, organisational and environmental factors in the pathogenesis and spread of brucellosis. Sudden changes in climatic conditions, in particular temperature fluctuations and increased humidity in the spring and summer, contributed to the activation of the pathogen and the development of outbreaks

of the disease. Additional risk factors were a decrease in the immune status of animals due to helminthiasis and metabolic disorders.

The results of the study substantiated the need for a comprehensive approach to the diagnosis and prevention of brucellosis, including timely laboratory confirmation of the diagnosis, differentiation from similar diseases, and the implementation of vaccination programmes taking into account the specifics of regional epizootic conditions. An important element of anti-epizootic measures is also the sanitary treatment of infection foci and the introduction of quarantine restrictions to prevent the spread of the disease. In the future, it is advisable to deepen the study of the molecular-genetic characteristics of the pathogen and the factors affecting its virulence, as well as to monitor the effectiveness of existing vaccines in different climatic zones. Particular attention should be paid to the development of integrated epizootic control systems and improving farmers' knowledge of animal hygiene, which will help to reduce morbidity and economic losses in sheep breeding.

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