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MICROBIOLOGY

Biosecurity practices associated with bovine viral diarrhea virus infection in dairy herds in Brazil

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ABSTRACT: This study determined the association between biosecurity practices and the status of bovine viral diarrhea virus (BVDV) in dairy production systems. Approximately 280 herds were screened for BVDV virus detection. Following the screening, 68 herds were selected to identify individual BVDV PI animals using an ear notch biopsy and ELISA-antigen. All offspring of the last generation were tested, and the maternal lineage of positive cases was examined. A questionnaire on BVDV biological risk assessment was completed. A multiple correspondence analysis (MCA) was conducted to determine the association between herds with or without BVDV circulation and biosecurity practices. The MCA revealed that farms with virus circulation lacked knowledge about the disease and wrongly perceived their herds as protected, while farms without virus circulation were aware of the disease but considered their herds unprotected. Vaccination practices differed between positive and negative herds, with positive herds using vaccines only for reproductive diseases and negative herds vaccinating for respiratory and reproductive issues. Biosecurity practices such as frequent visitation, contact between animals of different ages, and annual introduction of new animals were linked to viral circulation, while virus-free herds implemented measures like controlled visitation, no contact between different age groups, and quarantine. Lastly, herds with virus circulation acquired pregnant females without prior testing. This study emphasized the crucial role of biosecurity practices in controlling BVDV in dairy herds. It highlighted the sharper risk perception and better application of biosecurity practices in negative herds compared to BVDV-positive herds.
Key words: preventive measures, risk management, biosafety, bovine viral diarrhea virus.

Práticas de biosseguridade associadas com infecção do vírus da diarreia viral bovina em rebanhos leiteiros no Brasil

RESUMO: Esta pesquisa teve como objetivo associar as práticas de biosseguridade com a circulação do vírus BVDV em sistemas de produção leiteiros. Inicialmente, 270-290 sistemas foram selecionados para detecção do vírus entre janeiro de 2020 e janeiro de 2023 por meio da técnica de Reação em Cadeia da Polimerase em Tempo Real (RT-qPCR em tempo real). Após essa etapa, 68 rebanhos foram selecionados para identificação de animais persistentemente infectados (PI) por meio da análise individual do fragmento auricular usando o método ELISA-antígeno. Todos os animais nascidos na última geração, fêmeas que deram à luz bezerros machos ou natimortos, foram testados incluindo a pesquisa na geração de mães e avós maternas em casos positivos. Um questionário de análise de risco biológico para o BVDV foi aplicado a esse grupo de fazendas. Para identificar associações entre rebanhos com ou sem circulação do BVDV e práticas de biosseguridade, foi usada a análise de correspondência múltipla (MCA). A análise mostrou associação entre a falta de conhecimento sobre a doença e a percepção de que o rebanho está protegido contra o BVDV em fazendas com circulação do vírus. Os rebanhos positivos para Diarreia Viral Bovina reportaram o uso vacinas apenas para doenças reprodutivas, enquanto os rebanhos negativos reportaram o uso vacinas para problemas respiratórios e reprodutivos, sugerindo possíveis diferenças nos protocolos de vacinação e na eficácia das vacinas no Brasil. Práticas de biosseguridade como visitas frequentes, contato entre animais de diferentes idades e a introdução anual de novos animais, foram associadas à circulação viral, enquanto os rebanhos livres de vírus adotaram medidas como políticas de visitação e controle, ausência de contato entre os bovinos de diferentes idades e quarentena. Além disso, os sistemas de produção positivos adquiriram fêmeas prenhes sem testar o animal previamente. Assim, este estudo destaca aspectos essenciais para o controle do vírus da Diarreia Viral Bovina em rebanhos leiteiros, identificando que gestores de rebanhos negativos possuem maior percepção de risco associada à implementação de práticas de biosseguridade, em contraste comos rebanhos positivos para o BVDV.
Palavras-chave: medidas preventivas, gerenciamento de riscos, biossegurança, virus da diarreia viral bovina.

INTRODUCTION

Controlling infectious diseases in dairy production systems is essential to ensure herd health, especially from agents responsible for productive and reproductive losses, such as bovine viral diarrhea virus (BVDV). Virus presence can cause significant direct losses, including mortality, morbidity, early slaughter, stillbirths, abortions, and reinfection, as well as indirect losses from the costs of implementing control

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programs (PINIOR & FIRTH, 2017). RICHTER et al. 2017 determined direct losses ranging from 0.5 to 687.80 USD per animal. In Germany, on average, estimated direct costs exceed 110 million Euros per year (GETHMANN et al., 2019).

BVDV is an enveloped RNA virus belonging to the genus *Pestivirus* of the *Flaviviridae* family, and has great genetic variability (BVDV 1, BVDV 2, HoBi-like Pestivirus, Border Disease Virus, and Classical Swine Fever Virus). It is the causative agent of BVD and is distributed worldwide. The clinical manifestations of BVDV infection depend on the immune status, gestational age, and concomitant infections with other pathogens. BVDV can overcome the placental barrier when it infects a pregnant female, and when it infects during the gestational period, there is immunotolerance to the virus, resulting in persistently infected (PI) calves. PI cattle eliminate the virus from nasal ocular secretions and the virus can be found in various animal tissues. If infection occurs in cattle with immune capacity to respond to the virus, acute or transient infection occurs mainly through muzzle-to-muzzle contact (direct contact) (WALZ et al., 2010). Other consequences include problems with the embryo or fetus, lower conception rates at first service, embryonic loss, infertility, teratogenesis, and miscarriages (GRISSETT et al., 2015). Postnatal cross-transmissions result in transient infections. Transiently infected animals exhibit fever, nasal discharge, enteritis, and leukopenia; they also have a higher frequency of diarrhea (33%) and a greater chance of developing bovine respiratory disease. Bovine Viral Diarrhea viruses can also remain lodged in ovarian tissues for long periods after an acute infection (BASQUEIRA et al., 2020; GIVENS & MARLEY, 2008; GROOMS et al., 1998).

The cellular and humoral responses in BVDV-affected cattle become impaired. The virus affects two of the main leukocyte types, neutrophils and macrophages, inducing neutrophil apoptosis, reducing the expression of CD18 and L-selectin, and reducing macrophage activity, depending on the BVDV strain. Humoral immunosuppression occurs shortly after viral infection, mainly at the IgG level (ABDELSALAM et al., 2020; THAKUR et al., 2020; RAJPUT et al., 2020).

Control and eradication programs for BVDV using a combination of diagnostic tests and PI animal disposal have been successful in countries such as Finland, Sweden, Norway, Denmark, Austria, and Switzerland. The European Union has national plans to eradicate the virus, and Germany, Austria, Scotland, and Ireland are adept at achieving this goal. In other countries such as England and France, adherence is voluntary. Control and eradication programs for BVDV must include three pillars: detection of PI animals, vaccination, and implementation of biosecurity practices. However, in Brazil, there is no mandatory or specific control or eradication program for BVDV.

The approach chosen to implement a biosecurity program can be based on the segmentation of biosecurity into internal and external, or bio-compartments (RENAULT et al., 2021). In both programs, the measures adopted must consider the transmission routes of the pathogen. GATES et al. (2013) reported the importance of requiring herds infected with BVDV to adopt biosecurity measures to prevent the disease spread. CARDWELL et al. (2016) reported that the introduction of a biosecurity program monitored for to 2–3 years tended to reduce the risk of introducing and spreading biological agents in herds.

This study aimed to identify associations between biosecurity practices and BVDV circulation in dairy herds in southern Brazil.

MATERIALS AND METHODS

Target *population*

BVDV was tested in dairy herds associated with a dairy cooperative in the state of Paraná in southern Brazil. The properties were located within 113 km of the cooperative's headquarters in Carambeí. The herds were characterized as being predominantly intensive, with an average daily production per herd of 4,454 liters, 183 lactating cows, 31 liters/cow, DIM of 197, and somatic cell count of 218,000 cells/mL. Complete data on the herd characterizations are in the online repository (Supplementary File S1, https://doi.org/10.5281/zenodo.10034748).

BVDV testing was performed at both herd and individual levels. In summary, in the first stage 270–290 production systems were selected from January 2020 to January 2023 for BVDV detection in the expansion tank using a real-time polymerase chain reaction (RT-qPCR) (IDEXX RealPCR BVDV RNA Test; IDEXX, Westbrook, ME, USA), exceeding 2,900 tests.

Of these, 68 herds were selected for PI testing, detected using ear biopsy and individual analysis from an enzyme-linked immunosorbent assay (ELISA)-antigen (IDEXX BVDV Ag/Serum Plus Test). To determine the presence of PI animals in the herd, all animals from the last generation, including females that gave birth to males or stillbirths, were tested. If the results were positive, the tests were repeated between days 21 and 30. Animals that tested positive in two consecutive tests were classified as PI, and those that tested positive in only one test were classified as transiently infected. The mothers and grandmothers of the PI animals were also tested following the same protocol.

The owners involved in the study signed a form stating they were responsible for the disposal of the PI animals once diagnosis was confirmed.

The herds considered negative in both the RT-qPCR and ELISA antigen tests had negative results in all the tests carried out during this period. Herds that had at least one positive result in both RT-qPCR and ELISA antigen tests were considered positive. The number of tests per herd during the period varied from 1–13 for the RT-qPCR and 1–12 for the ELISA-antigen test. As a result, 24 farms were negative for both tests using the RT-qPCR and/or ELISA-antigen; 13 farms were positive only for the RT-qPCR; 11 farms were positive only for ELISA-antigen; and 14 farms were positive in both tests.

The questionnaire for assessing biological risk in herds included questions related to general biosecurity practices and BVDV, which are available in Supplementary File S2. (https://doi.org/10.5281/zenodo.10027096).

Statistical analyses

Data were tabulated in Microsoft Excel (Microsoft Corp., Redmond, WA, USA) and descriptive statistics were analyzed using Statistical Analysis System (SAS) software version 9.4 (SAS Institute Inc. NC, USA).

A multiple correspondence analysis (MCA) was used to verify the association between herds with or without BVDV circulation and biosecurity practices using the JMP program integrated into SAS (version 17, premium). The MCA shows the grouping of variables inserted as dimensions (dimensions 1, 2, 3, ... n) in ascending order, according to the percentage of variability explained. From the inertia value, it was also possible to verify each variable's contribution to the dimensions (DENIS-ROBICHAUD et al., 2019).

RESULTS

Biosecurity practices and BVDV viral circulation

The biosecurity measures in the dairy production systems evaluated included access control for people, animals, vehicles, equipment; animal health management; reproductive management; factors related to calving; vaccination practices; cleaning and disinfection protocols; and producers' perceptions of biological risk. The distribution of biosecurity practices implemented in positive and negative herds can be assessed on https://doi.org/10.5281/zenodo.10180690. In general, the frequency of biosecurity practices in negative herds was similar to that in positive herds regardless of the test used in the MCA. Regarding the entry of disposal trucks into the properties, the clean areas for ELISA antigen-negative herds had a higher rate of prohibition of vehicle entries of this type.

MCA between qPCR and ELISA-antigen test results and biosecurity measures

The biosecurity measures analyzed were grouped according to producer perceptions for the risk of reproductive viruses, and the other answers were grouped into blocks: access control for people, animals, and vehicles, in addition to blocks made up of factors related to calving, health management, reproductive management, cleaning and disinfection process (CDP), conservation of vaccines and medicines, and good vaccination practices. This division was employed to better visualize the data in the MCA.

The MCA generated three dimensions (d1 = 58.12%, d2 = 37.41%, d3 = 4.47) for the variables of BVDV risk perception and infection status of the production systems, explaining 100% of the data variability. In summary, the risk perception of those responsible for positive herds was worse than that of those responsible for negative herds. For positive herds, there was a correlation between lack of knowledge about BVDV and attitude awareness in the case of positive animals. In addition, there was an association between positive infection status and producers' perception that their herds were protected against BVDV, they also perceived that the animals most affected by the reproductive virus were lactating cows. In contrast, managers responsible for negative herds were aware of the disease and considered their herds unprotected; for them, heifers were the category most predisposed to BVDV (Figure 1).

Vaccination to prevent reproductive diseases was associated with BVDV-positive herds (d1: 78.7%, d2: 20.1%, d3: 1.19%). The managers did not report the type of vaccine (inactivated or modified live) administered. Negative herds were associated with the use of inactivated dual-purpose vaccines (reproductive and respiratory vaccines). The type of adjuvant presents in the immunizers had little correspondence with the positive status of the herds, mainly because almost all farms administered vaccines containing aluminum hydroxide (Figure 2).

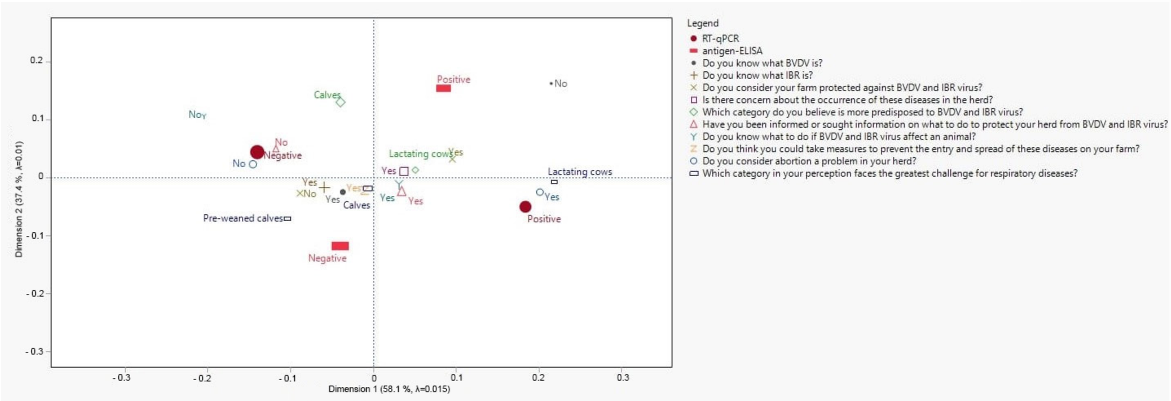

Figure 1 - Multiple correspondence analysis between the risk perception of producers in the Campos Gerais region of Paraná and bovine viral diarrhea virus (BVDV) viral circulation in their herds.

Regarding biosecurity practices to prevent the entry, dissemination, and/or exit of viral agents, MCA between access control and health status showed a greater number of weekly visitors for positive herds than for negative herds, and negative herds had visitation policies (Figure 3). In this analysis, the first two dimensions explained more than 98% of the data variability (d1 = 60.90, d2 = 37.31, d3 = 1.79), in which the first dimension was characterized by the number of weekly visitors (x-axis) and the second by whether employees avoided contact with cattle outside the property (y-axis). Regarding animal traffic, herds with viral circulation had contact with other categories of cattle; there was an annual introduction of new animals, and the same animals left and re-entered the property for external management three to six times a year. Negative herds were associated with the absence of contact with animals of the same species of different ages, quarterly transportation of the same animals for external management, ease of quarantine, and brucellosis and tuberculosis testing (d1 = 68%, d2 = 27.5%, d3 = 4.5%) (Figure 4). Only one farm tested its animals for brucellosis, tuberculosis, and BVD. The main animal species living on farms, apart from cattle, were dogs, cats, horses, and deer (d1 = 83.4%, d2 = 13.4%, d3 = 3.2%) (Figure 5). However, the correspondence obtained by the MCA between other animal species and viral circulation was not as evident. Four questions assessed vehicle and equipment traffic control; positive herds did not have tractors/equipment for towing animals, a specific parking space, or ban entry for waste disposal trucks.

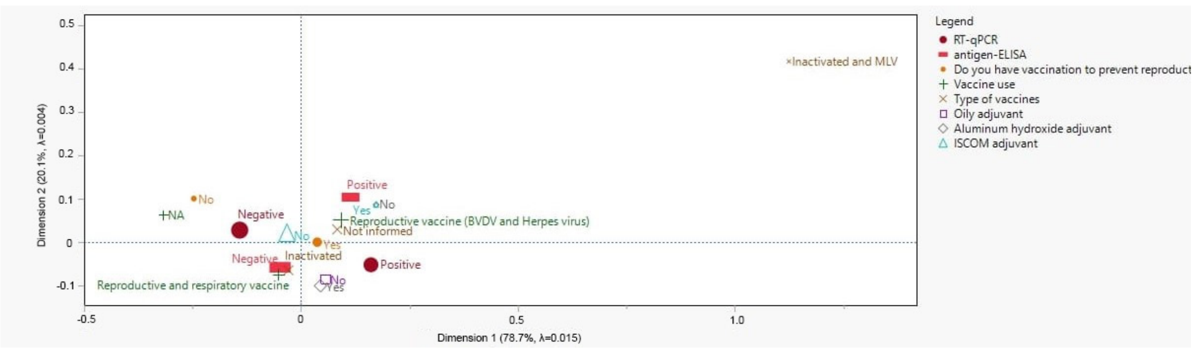

Figure 2 - Multiple Correspondence Analysis between vaccine and drug conservation measures and BVDV viral circulation on dairy farms in Brazil.

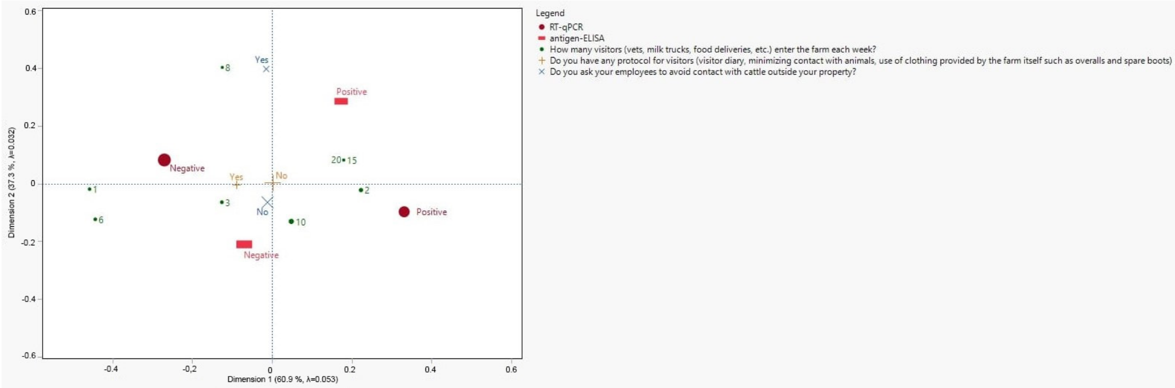

Figure 3 - Multiple Correspondence Analysis between practices for controlling human access and BVDV viral circulation on dairy farms in Brazil.

In contrast, negative herds did have specific parking space and ban entry for waste disposal trucks (d1 = 83.5%, d2 = 12.8%, d3 = 3.7%) (Figure 6).

Considering factors related to the calving of bovine females, positive herds were associated with the acquisition of pregnant females that calved on pasture. The abortions occurred bimonthly and quarterly, and the aborted fetus was disposed of in the field (Figure 7). Negative herds were associated with not purchasing pregnant females or giving birth in maternity paddocks and compost barns. Monthly and annual abortion frequencies were also correlated. When abortions occurred, diagnostic tests were conducted on the animals and the aborted fetus was buried. Not separating the aborting female from the herd, not carrying out laboratory tests on the female and aborted material, and not assisting cows and heifers in giving birth did not correlate with serological status. (d1 = 70.2%, d2 = 27.8%, d3 = 1.92%).

Positive herds corresponded to difficulties in isolating sick or quarantined animals from healthy animals. The drugs used to treat respiratory diseases associated with a positive status were florfenicol and tilmicosin (Figure 8). Negative herds corresponded to the ease of isolating sick animals, isolation/ quarantine sites not being close to healthy animal facilities, and young and adult cattle not being monitored for respiratory diseases. In this group, the two main substances used to treat respiratory diseases were tilmicosin and enrofloxacin (d1 = 71.7%, d2 = 26.9%, d3 = 1.37).

Regarding reproductive management, BVDV-positive herds corresponded with females eligible for reproduction between 11 and 14 months

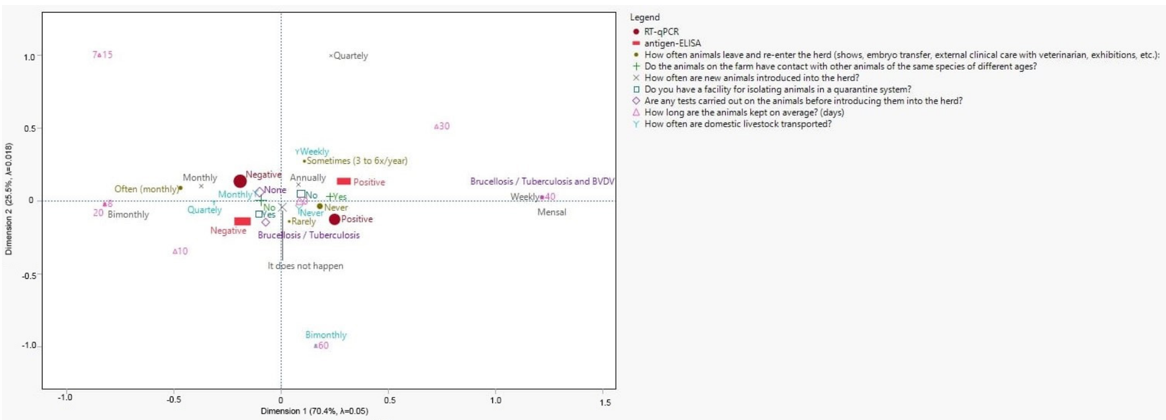

Figure 4 - Multiple Correspondence Analysis between animal access control practices and BVDV viral circulation on Dairy farms in Brazil.

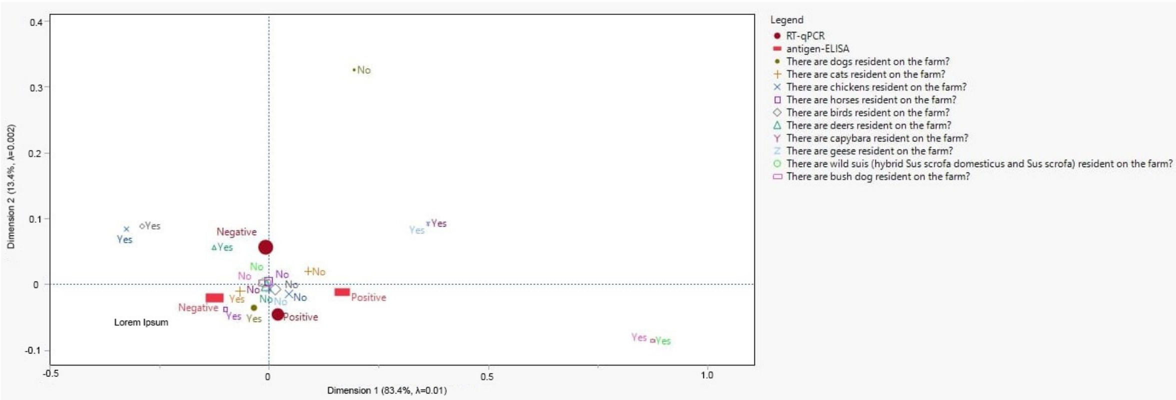

Figure 5 - Multiple Correspondence Analysis between other animal species present in the herds and BVDV viral circulation in dairy systems.

of age, bulls not used for reproduction, purchase of pregnant females, and metritis as the main cause for cows being discarded (d1 = 76.82%, d2 = 18.71%, d3 = 4.47%). For the negative herds, the age of females suitable for reproduction ranged from 10 to 26 months, and managers either purchased pregnant animals or did not. The main reasons for excluding cows from negative herds were low milk production and reproductive disorders (Figure 9).

The procedures for cleaning and disinfecting calf cages in the rearing phase and employees washing hands before handling them was absent in the positive herds (Figure 10). Both practices were observed for the negative herds (d1 = 76.82%, d2 = 22.11%, d3 = 1.07%). The absence of a CDP at the calving site after an abortion was not associated with viral circulation. Regarding the management of waste produced in the dairy system, farms with positive herds managed both bedding and manure, or manure only (Figure 11). Manure is used for composting, farming, and biodigestion. When present, animal bedding buried, or lime (lime hydroxide) was included. Farms with negative herds managed waste using septic tanks and solid-liquid separation, with solid waste designated for crops. In farms with positive herds, carcasses were disposed in crops or buried; in negative herds, they were left in the pasture, ditches, or far from the property (d1 = 66.1%, d2 = 30.8%, d3 = 3.1%).

DISCUSSION

This study evaluated the association between herd infection status and biosecurity practices related to the control of BVDV.

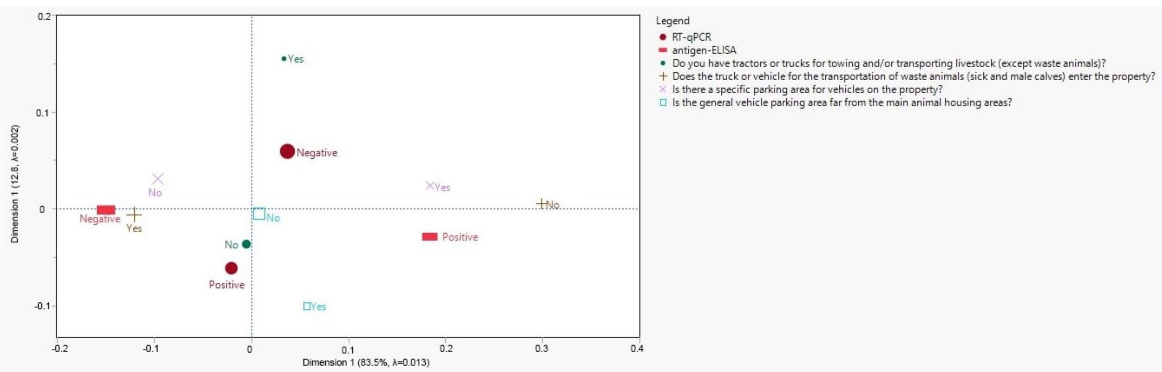

Figure 6 - Multiple Correspondence Analysis between biosecurity measures aimed to control vehicle traffic and BVDV viral circulation on dairy farms in Brazil.

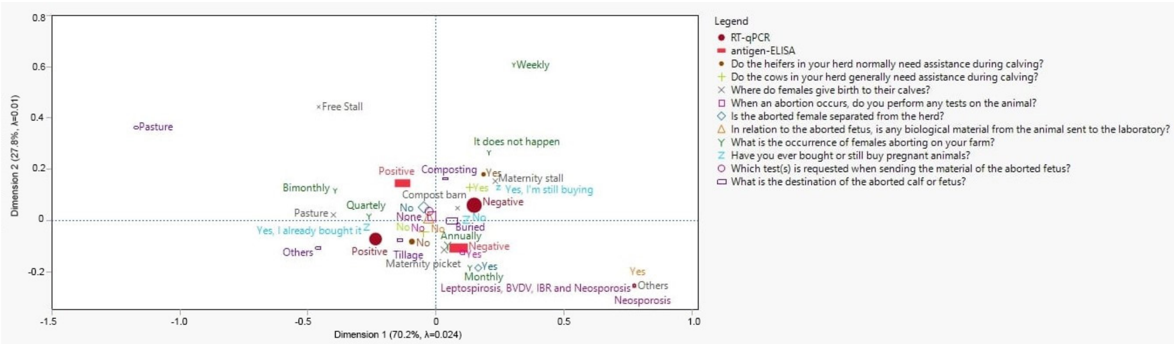

Figure 7 - Multiple Correspondence Analysis between factors related to the calving of bovine females and BVDV viral circulation on dairy farms in Brazil.

Biological risk assessments include an assessment of producers' perceptions of risk. We identified contradictory responses in associations between BVDV presence in herds and risk perception. Positive herds were associated with concern about the disease in the herd, and managers responded that they had been informed about the BVDV previously. At the same time, they considered their farm to be protected from the virus and were unaware of BVD. Similarly, a previous study by GATES et al. (2013) reported that 25% of the producers with seropositive herds recognized that their herds were infected with BVDV. Producers' risk perceptions comprise life experiences, culture, education, and age. These factors influence their willingness to take risks when encountering health challenges and adherence to biosecurity measures (RENAULT et al., 2020).

We observed a great opportunity to improve vaccination protocols in herds; this practice represents one of the pillars for a BVDV control and eradication program. In our study, positive herds were associated with single-purpose inactivated vaccines, indicating that they were only protected against reproductive diseases. In contrast, negative herds were associated with dual-purpose vaccines, which suggests the early adoption of vaccination protocols at a young age on farms that use formulations designed to prevent respiratory and reproductive diseases with a greater anamnestic response and future protection. The fact that herds were positive for the virus, even with vaccination practices in place, may suggest the low efficiency of commercial vaccines used in Brazil and/or poor vaccination practices. Several research groups (BACCILI et al., 2019; ARENHART et al., 2008) have questioned the effectiveness of inactivated BVDV vaccines in Brazil. NISKANEN & LINDBERG (2003) conducted a study on a PI-positive herd and observed horizontal transmission of BVDV during unhygienic vaccination management. PINIOR et al. (2019) showed that the implementation of vaccination and biosecurity measures is associated with a reduction in milk production loss. In herds that

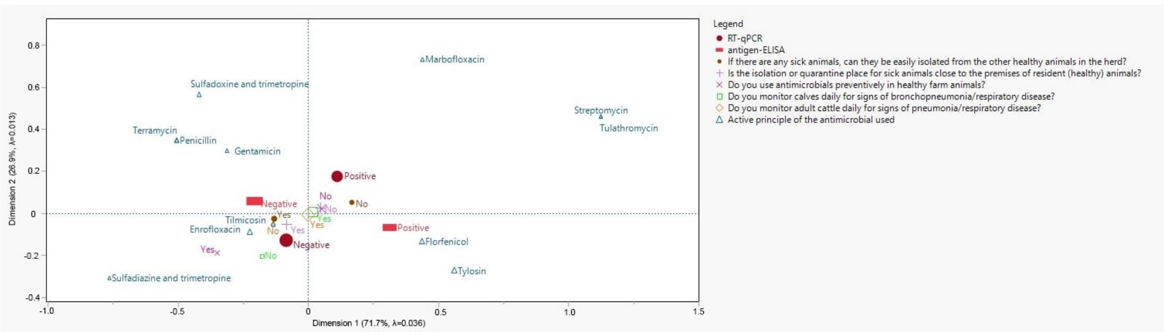

Figure 8 - Multiple Correspondence Analysis between bovine animal health management and BVDV viral circulation on dairy farms.

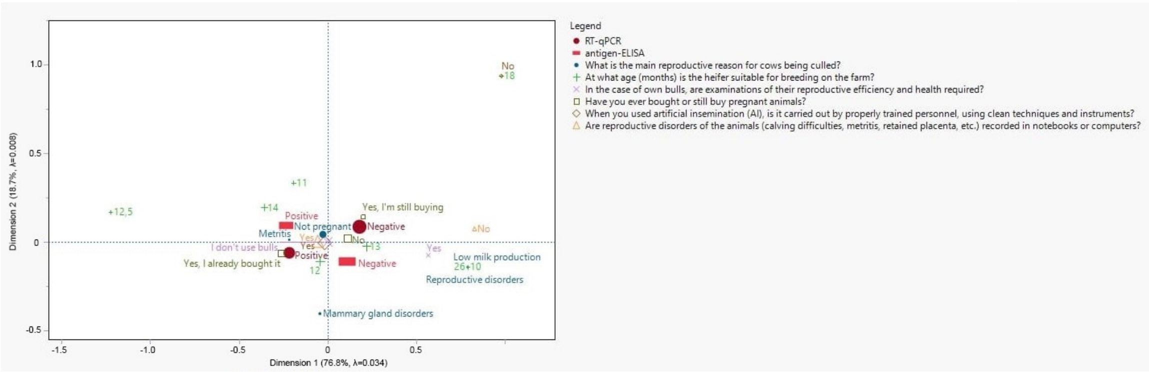

Figure 9 - Multiple correspondence analysis between reproductive management and BVDV viral circulation on dairy farms.

implemented biosecurity measures, losses ranged from 8% to 12%, whereas in herds without these measures, losses ranged from 28% to 29%.

Biosecurity measures that target the control of human and vehicle traffic are intended to prevent agents from entering herds. Positive herds had a greater variation in the number of visitors and did not have tractors or trucks to transport the animals, whereas negative herds had protocols for visitors and allowed trucks to transport discarded animals to clean areas. Practices such as providing visitors with protective clothing and boots, restricting the drivers of vehicles transporting animals from encountering the farm's resident animals, and ensuring that trucks transporting animals for calf culling arrive empty were the measures with the greatest impact on the likelihood of infection through indirect contact for the majority of farms in a study by BENAVIDES et al. (2020). Herds with virus circulation had an annual introduction of new animals, and the same animals left and re-entered the property for external management three–six times per year. BENAVIDES et al. (2020) listed actions that contributed 12% of annual introduction to BVD, such as the acquisition and entry of new animals, raising heifers outside the property, sending cattle out to events, sharing vehicles to transport animals, and the absence of a CDP when sharing occurred.

In our study, the main calving sites were the maternity paddocks, with an association between RT-qPCR-positive herds and females calving in free stalls and compost barns. RYPULA et al. (2020) revealed that differences in BVDV prevalence were linked, among other factors, to grazing practices, which are lower in extensive systems. The BVDV is enveloped, which makes it more sensitive to disinfectants and sunlight.

Considering the practices in the investigated dairy farms, we identified five risk factors for BVD: herd size, herd type [dairy/beef], participation in agricultural events/shows, introduction of cattle, presence of pasture and grazing animals, and contact with them.

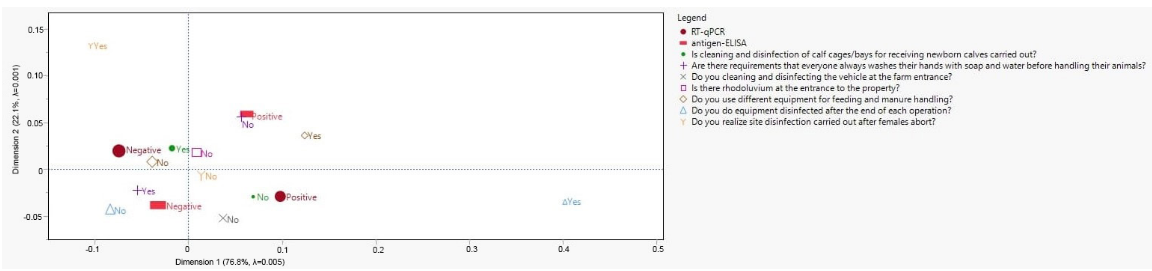

Figure 10 - Multiple correspondence analysis between cleaning, disinfection procedures, and BVDV viral circulation on dairy farms.

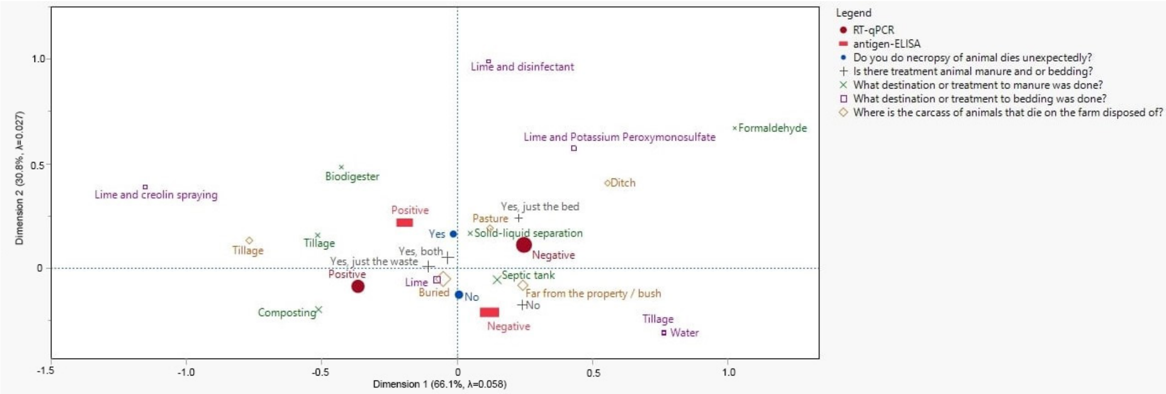

Figure 11 - Multiple correspondence analysis between cleaning and disinfection procedures and the disposal of bedding, carcasses, and animal waste and BVDV viral circulation on dairy farms.

The two main drugs associated with the BVDV-positive herds were florfenicol and tylosin. According to the European Medicines Agency (EMA), the conscious use of antimicrobials for human and animal health can reduce the risk of antibacterial resistance. Researchers have categorized antimicrobials into four classes (A, B, C, and D) based on their effects on public health and bacterial resistance. Therefore, the two drugs most used by the producers in our study fell into category C, which comprises antimicrobials that should be used with caution and only considered in treatment protocols when there is no effective substitute in category D. Negative herds were strongly associated with the use of enrofloxacin and tilmicosin, which belong to classes B and C. Despite their herds being negative for BVDV, this group of producers should be warned that the use of enrofloxacin should be restricted because it is extremely harmful in human medicine, and other alternatives in categories C and D should be considered.

From the perspective of reproductive management, artificial insemination was the only reproductive technique used, although 7.24% of respondents reported requesting tests to assess the efficiency and reproductive health of bulls when they use them. Regarding biological risk, farms that have bulls in their herds for breeding have a higher, but not significant, chance of BVDV infection [OR = 1.16, 95% CI 0.90–1.49] (WILLIAMS & WINDEN, 2014). The main reason for culling cows in positive herds was metritis, whereas in negative herds it was low milk production and reproductive diseases. BASQUEIRA et al. (2020) found that PI cows were 1.6 times more likely to develop metritis than non-PI cows. In addition, all lactating PI cows had at least one postpartum illness, including mammary gland edema, retained placenta, ketosis, hypocalcemia, and clinical mastitis.

To control the spread of the virus and other pathogens within and between herds, hygiene and disinfection procedures are essential to ensure animal health and welfare, especially in intensive systems where animal density and productivity increase infection risk. Adapting a CDP according to the production system can help reduce pathogen levels and prevent or interrupt the disease cycle (CVM, 2023). In our study, we identified the absences of a CDP in stalls and cages in calving areas, hand hygiene before handling animals, and a CDP after abortion in positive herds. NISKANEN & LINDBERG (2003) reported that two of three calves tested positive for BVDV after being housed in a pen shortly after the removal of a PI calf without the pen being cleaned and disinfected. In addition to the importance of BVDV, MEGAHED et al. (2022) identified measures such as postpartum cleaning and disinfection procedures, abortion history, and sharing equipment as potential risk factors for *Brucella* infection.

From an environmental point of view, waste management in dairy production systems must be concerned with its impact on water quality, odor emissions, and pollutants such as ammonia. Animal waste on positive herd farms consists of compost, agriculture, or biodigesters. Negative herds manage their waste using septic tanks and solid-liquid separation, where the solids are destined for farming. Manure application to the soil at acceptable fertilizer levels for crops produced on farms through the transport or pumping of washed manure effluents through irrigation systems is the basis of most systems. In contrast, the energy generated from biodigesters reduces the emissions of methane and odorous components (VAN HORN et al., 1994).

Carcasses from properties with BVDV viral circulation were handled and disposed of either by plowing or burying them, whereas those without the virus were left in the pasture, ditches, or far from the property. Issues such as soil pollution, water contamination through groundwater, and odors have been discussed by VAN HORN et al. (1994). XU et al. (2009) determined that the rapid decomposition of dry matter from bovine carcasses reduced the viability of cells from harmful agents through the composting process, even for outbreaks of infectious diseases caused by *Escherichia coli* O157:H7, *Campylobacter jejuni*, and Newcastle disease virus, which were inoculated for during the study. Thus, proper composting can be an alternative for adequate disposal of carcasses within the animal system.

Finally, we identified opportunities for the dairy systems in the study to adopt and adapt existing biosecurity practices for both animal health and environmental aspects. Among the main animal-focused measures were access control, quarantine, testing animals, proper carcass disposal, and use of antimicrobials.

CONCLUSION

This study identified specific biosecurity practices of BVDV-positive and negative dairy herds. These practices included having more visitors, allowing contact between cattle of different ages, and sharing tractors and equipment between farms. BVD-negative herds had protective practices to mitigate the main risk factors of the disease. However, these measures could be more widespread in the dairy industry in Brazil. This information will help those responsible for the farms prioritize measures that should be adopted in their herds. Benchmarking tools between production systems could encourage herd facilities to adopt these practices. Further studies are needed to link the economic and health impacts of herds to biosecurity.

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DECLARATION OF CONFLICT OF INTEREST

The authors declare that this study was conducted in the absence of any commercial or financial relationships that could be construed as potential conflicts of interest.

AUTHORS' CONTRIBUTIONS

JSF: conceptualization, data curation, writing-original draft, writing-review and editing; BSN: conceptualization, writing-original draft, writing-review and editing; CCB: conceptualization, data curation, supervision, formal analysis, writing-original draft, writing-review and editing; FKV: conceptualization, data curation, writing-original draft, writing-review and editing; LMS: conceptualization, data curation, writing-original draft, writing-review and editing; TI: conceptualization, data curation, writing-original draft, writing-review and editing; JTP: conceptualization, data curation, writing-original draft, writing-review and editing; VG: conceptualization, data curation, supervision, writing-original draft, writing-review and editing.

BIOETHICS AND BIOSECURITY COMMITTEE APPROVAL

This study was approved by the Ethics Committee on Animal Use of the Faculty of Veterinary Medicine and Animal Science of the Universidade de São Paulo (CEUA/FMVZ) (protocol number: 1288180520) and for humans (Platform Brazil number: 37108020.2.1001.5390).

SUPPLEMENTARY MATERIAL

Supplementary Materials for this article can be read online:
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