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# The Example of Çankırı Province in Combating Brucellosis in Cattle

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**ABSTRACT**

Brucellosis in bovines is an infectious disease that leads to significant health, productivity, and economic losses. It is also a zoonotic disease, posing a serious threat to public health. This disease is widespread in Türkiye as well as globally. Vaccination is a crucial and cost-effective measure, with the most commonly utilized vaccine strains worldwide for bovine brucellosis control being *B. abortus* S19 and RB51. In this study, the results of the intensive efforts to combat bovine brucellosis between 2011 and 2015 in Çankırı province, as well as the results of the mass *B. abortus* S19 conjunctival vaccination campaigns carried out within the scope of the Ministry project in 2012-2013, are evaluated. Within the scope of the research, retrospective data obtained from various public institutions affiliated with the ministry were evaluated. The results of disease control, eradication and vaccination studies carried out in the field were evaluated epidemiologically within the framework of legal regulations. As a result of close monitoring of bovine brucellosis outbreaks and mass conjunctival vaccination in Çankırı province, the number of outbreaks has decreased more than four times compared to the average in Türkiye. Additionally, there is a 95-fold reduction in the number of animals compensated for the disease and a more than 200-fold decrease in the total amount of compensation paid. This study examines the efforts to control bovine brucellosis in Çankırı province, particularly focusing on mass conjunctival vaccination campaigns using the *B. abortus* S19 vaccine strain. Given the conditions in Türkiye, the *B. abortus* S19 vaccine strain is currently deemed the most effective for preventing brucellosis in bovines. Ultimately, to attain the planned objectives in bovine brucellosis control in Türkiye, the study concludes that vaccination during a suitable period, which does not pose a risk to pregnancy and ensures robust herd immunity, is the optimal approach.

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## INTRODUCTION

Bovine brucellosis is a contagious and infectious disease that is usually caused by *Brucella abortus* (*B. abortus*) and results in abortion, premature calf birth, infertility, and productivity losses in cattle (Peker et al., 2010; Inlamea et al., 2016; Khurana et al., 2021; Rahimnahal et al., 2023). Bovine brucellosis, which is also a zoonotic disease, is also an important cause of serious public health problems and economic losses (Güzelant et al., 2009; Ulaş et al., 2012; Khurana et al., 2021; Rahimnahal et al., 2023). *B. abortus* is one of the most common agents in bovine and has been classified as a category (B) pathogen with the potential to be used as a biological weapon (Peker et al., 2010; Khurana et al., 2021). Except for brucellosis-free countries, it is widespread worldwide and its prevalence is high especially in developing countries in the Middle East, Central Asia, and the Mediterranean (Güzelant et al., 2009; Peker et al., 2010; Inlamea et al., 2016; Kepenek Kurt et al., 2021). Brucellosis in Türkiye is particularly prevalent in the Central, Eastern, and Southeastern Anatolia Regions. It is a zoonotic disease that is widespread in our country and persists as an ongoing public health concern (Demir and Orhan, 2012; Sayar et al., 2019; Kepenek Kurt et al., 2021).

Brucellosis is recognized by the World Health Organization (WHO) and the Food and Agriculture Organization (FAO) as the most common zoonotic disease worldwide (Khurana et al., 2021; de Oliveira et al., 2022). Laine et al. (2023) reported that the annual incidence of human brucellosis worldwide is approximately 1.6-2.1 million new cases, according to their calculations (Laine et al., 2023). Infected animals are the main source of infection in humans. Transmission to humans usually occurs through the consumption of unpasteurized milk or dairy products obtained from infected animals (especially raw milk, fresh cheese, butter, etc.) (Ulaş et al., 2012; Sayar et al., 2019; Kepenek Kurt et al., 2021). The disease is more commonly observed in rural areas among livestock breeders, veterinary health workers, butchers, slaughterhouse and milkers, and scientists. Bovine brucellosis is also referred to as contagious abortion or Bang's disease (Peker et al., 2010; Khurana et al., 2021). The primary clinical symptoms observed in humans include shaking, high fever, weakness, as well as muscle and joint pains (Demir and Orhan, 2012). An effective vaccine has not yet been developed to protect people from brucellosis (Güzelant et al., 2009). Currently, vaccinating animals, pasteurizing milk, and taking measures to prevent contact with infected animal products and materials are the most effective methods for preventing transmission and protecting humans from brucellosis infection (Peker et al., 2010).

In addition to affecting public health, bovine brucellosis also affects the health of animals, reducing their productivity and causing significant economic losses in the livestock sector (Rahimnahal et al., 2023). The disease tends to spread rapidly within the infected herd, leading to a decrease in milk and meat production, as well as occurrences of abortions, stillbirths or premature births, and reproductive losses such as infertility (de Oliveira et al., 2022). The economic importance attributed to the disease is based on direct losses from abortions, loss of offspring, infertility, weight loss, decrease in milk production, and inhibition of trade in animals and their products (Dorneles et al. 2015). This disease also has negative socioeconomic effects, including a decrease in calf birth rate due to prolonged calving intervals, increased costs for purchasing re-breeding animals, diminished competitiveness in the market, treatment expenses, and loss of labor (Inlamea et al., 2016). It has been reported that the rate of abortion in susceptible herds varies between 30 and 80%. Brucella-positive animals are one of the major causes of the spread of brucellosis (Khurana et al., 2021). Due to the serious medical and socioeconomic consequences of the disease, it is important to prevent the transmission of the infection through vaccination of animals (Schurig et al., 2002).

Vaccination is the main measure to control bovine brucellosis. The most widely used vaccine strains in the world are *B. abortus* S19 and RB51 (Dorneles et al., 2015; de Oliveira et al., 2022). Both vaccines are effective in preventing abortion and infection, as well as providing long-term protection (Dorneles et al. 2015). The RB51 vaccine was developed in 1982 and is a rough rifampicin-resistant strain of *B. abortus* biovar 1 that does not express the O-side chain lipopolysaccharide in its membrane. Therefore, this vaccine does not stimulate antibodies detected by routine serological tests. There is no serological test available to detect RB51 infection (de Oliveira et al., 2022). Since the Brucella RB51 strain can pass into milk in animals, it can be transmitted to humans in this way and cause infections. Its protective effect in bovine is similar to that induced by strain S19. The most commonly used vaccine to prevent bovine brucellosis is *B. abortus* strain 19 (Schurig et al., 2002). This vaccine, which has been used since 1941, is a smooth-attenuated strain of *B. abortus* biovar 1 that induces an antibody response that is indistinguishable from that induced by infection (de Oliveira et al., 2022). While S19 is an older vaccine compared to RB51 and offers longer-lasting protection (over 10 years), it remains highly effective and cost-efficient, making it widely utilized (de Oliveira et al., 2022). Additionally, *B. abortus* S19 may offer cross-protection against *B. melitensis* (Khurana et al., 2021). Currently, the *B. abortus* S19 vaccine is considered the best for preventing brucellosis in cattle, but this vaccine, which contains dead and/or attenuated live strains, may have potential side effects. The most effective and cost-efficient method for controlling the disease is through rapid and accurate diagnosis coupled with appropriate animal vaccination programs (Rahimnahal et al., 2023).

This study discusses the efforts undertaken by the Ministry of Agriculture and Forestry (MoAF) and the Çankırı Provincial Directorate of Agriculture and Forestry to fight bovine brucellosis infection between 2011 and 2015, as well as the mass conjunctival vaccination studies conducted during 2012-2013. Our study is based on retrospective field data. The aim is to effectively monitor and control epidemics by creating strong herd immunity through *B. abortus* S19 conjunctival vaccination, and to produce successful results by preventing losses. For this purpose, the efficacy and results of *B. abortus* S19 conjunctival vaccine were evaluated in Çankırı province. The research results are intended to illuminate the ongoing efforts to fight the disease and to provide guidance for decision-makers.

## MATERIAL and METHOD

In this study, data from different institutions were used. The basic data of the research were taken from the records kept in the fight to control bovine brucellosis infection between 2011-2015 and the mass conjunctival vaccination studies applied with *B. abortus* S19 strain in 2012-2013 of MoAF, Çankırı Provincial Directorate of Agriculture and Forestry. According to this data, Çankırı province consists of 12 districts, including the central district. In these districts, there were a total of 9,327 cattle breeding premises in 2012-2013. The number of bovines in Çankırı province, which is the subject of the research in these years, are 133,010 and 130,658 heads, respectively. In these two years, it was determined that Çankırı province had 0.96% and 0.91% of Türkiye's overall bovine population, respectively.

The number of bovine brucellosis outbreaks in this period, the number of animals subject to conditions slaughtered due to the disease and the amount of compensation paid, the number of cattle within the scope of the *"Conjunctival Vaccine Control and Eradication of Brucella Project"*, the vaccination program and the vaccination figures applied were compiled retrospectively. In addition, the bovine brucellosis outbreak numbers reported by MoAF to the World Organization for Animal Health (WOAH) during this period were used.

Another data of the research is the total number of bovines throughout Türkiye. This data was taken from the 2012-2013 animal numbers of statistics kept by the Turkish Statistical Institute (TSI).

According to TSI data in 2012-2013, when mass conjunctival vaccination studies were carried out in the fight against the disease, the number of bovines in Türkiye was 13,914,912 and 14,415,257 heads, respectively.

Bovine brucellosis is designated as a notifiable disease in accordance with the *"Regulation on Notifiable Animal Diseases and Notification"* (Official Gazette (OG) dated 22.01.2011 and numbered 27,823) published under the auspices of Law No. 5996 on *"Veterinary Services, Plant Health, Food and Feed Law"* (OG dated 13.06.2010 and numbered 27,610). Currently, no treatment is provided for this disease. Furthermore, the *"Regulation on Fighting Brucellosis"* (OG dated 03.04.2009 and numbered 27,189) falls under the purview of Law No. 5996. Presently, efforts to combat this disease are conducted in accordance with the provisions outlined in this regulation, as well as the circular pertaining to the "*Fighting Animal Diseases and Control of Animal Movements"*, which undergoes annual renewal.

The *"Regulation on Compensated Animal Diseases and Compensation Rates"* (OG dated 14.01.2012 and numbered 28,173) and *"Regulation on Compensation for Animal Diseases"* (OG dated 06.03.2013 and numbered 28,579), under the scope of Law No. 5996 have been published. According to these regulations, if bovine animals are found to be infected with brucellosis through serological or bacteriological examination, they are required to be slaughtered subject to the condition or culled, with compensation paid at a rate of 9/10 of their appraised value.

The vaccine used for bovine brucellosis, under the trade name Brupen A ($5\text{-}10x10^9$ colony-forming units (CFU) for 1 dose of vaccine (50 µl)), was produced by the İstanbul/Pendik Veterinary Control Institute Directorate affiliated with the MoAF.

This epidemiological study involves analyzing data from the mentioned institutions, considering relevant legal regulations, and reviewing the outcomes of disease control and vaccination efforts conducted in the field.

Since retrospective data were used in this study, there was no need to obtain ethical permission. However, since bovine brucellosis is a notifiable disease, the necessary legal permission was obtained from MoAF, General Directorate of Food and Control (GDFC) to publish the research results. In addition, statements of consent were received from the relevant people for the images used in Figure 2.

## RESULTS

### *Outbreak Numbers*

Çankırı is a province located in the Central Anatolia Region. It is a settlement with restrictions for cattle breeding due to its geographical structure and pasture situation. Türkiye has a share of less than 1% of bovine assets. However, as can be seen detailed in Table, when we compare the number of bovine brucellosis outbreaks in Çankırı province and Türkiye in general, 57/486 (11.73%) in 2011 and 101/1.696 (5.96%) in 2012 were reported, respectively. In 2010, upon the notification of bovine brucellosis outbreaks density, on-site inspections were carried out in Çankırı Provincial Directorate of Agriculture and Forestry, Animal Health Branch Directorate and 11 District Directorates. Information on the subject was provided at the meetings attended by the provincial director, district directors, veterinary health workers and all other stakeholders. In these meetings, coordination was ensured between the teams and actions were taken in coordination with MoAF, GDFC. Within the possibilities, the personnel, document, and equipment deficiencies of the teams were eliminated. Existing equipment

was maintained and used in disinfection works. Since 2011, disease emergence and extinctions of transferred and newly notified outbreaks have been closely monitored. The premises where the disease is extinguished, and their surroundings are disinfected with sprayers. In 2012, a mass conjunctival vaccination program was initiated by MoAF within the scope of the *"Conjunctival Vaccine Control and Eradication of Brucella Project"*. As the details are given below, the bovine included in the program were vaccinated with a high percentage rate. As a result of all this fight against bovine brucellosis disease, there has been a significant decrease in the number of disease outbreaks over the years. In this context, in 2013, 2014 and 2015, the number of disease outbreaks reported in Çankırı was 30/1,319 (2.27%), 11/596 (1.85%) and 9/315 (2.85%), respectively, compared to Türkiye.

**Table**. Number of bovine brucellosis outbreaks in Türkiye and Çankırı province (MoAF, WOAH)

| YEARS | ÇANKIRI PROVINCE | TÜRKİYE |
|---|---|---|
| 2011 | 57 | 486 |
| 2012 | 101 | 1,696 |
| 2013 | 30 | 1,319 |
| 2014 | 11 | 596 |
| 2015 | 9 | 315 |

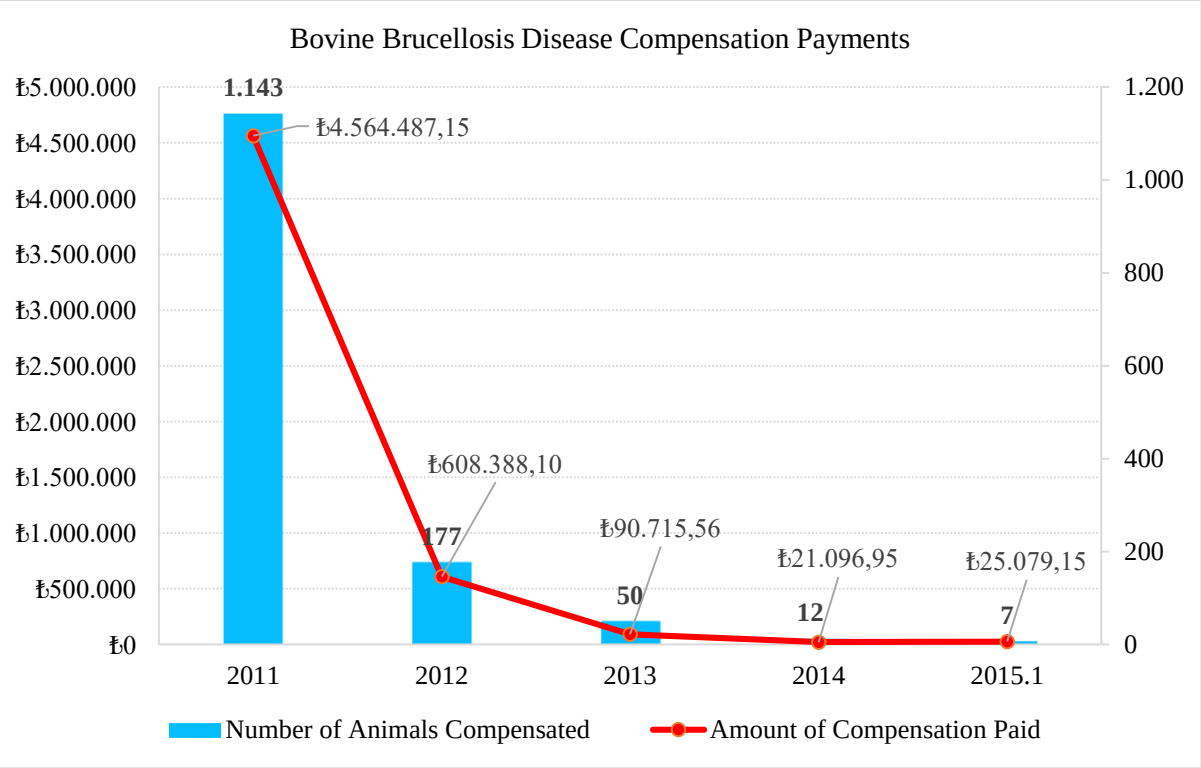
***Compensation Payments***

In this context, an intense fight has been carried out in Çankırı province since 2011. The animals that were detected as positive in the premises where the disease emergence was sent to conditional slaughter or culled. A compensation payment of 4,564,487.15 ₺ was made for 1,143 bovines, mainly due to bovine brucellosis in 2011. According to the information obtained, this figure corresponded to approximately quarter of the compensation paid for bovine brucellosis in Türkiye. As a result of the fight carried out and mass conjunctival vaccination studies, there has been a significant decrease in the number of animals culled or slaughtered subject to the condition and the amount of compensation paid, as the details can be seen in Figure 1. In 2012, 2013, 2014, and the first half of 2015, the number of bovines affected by bovine brucellosis and the corresponding compensation paid were 177 bovines (608,388.10 ₺), 50 bovines (90,715.56 ₺), 12 bovines (21,096.95 ₺), and 7 bovines (25,079.15 ₺), respectively. In the 4.5-year period (2011-2015.1) covered in this research, a total of 1,389 bovine were sent to conditional slaughter or culled due to bovine brucellosis. For these animals, a total of 5,309,766.91 ₺ compensation was paid to the breeders after deducting the roasting cost (Figure 1).

***Vaccination Program***

According to the final report of the project *"Eradication of Brucellosis and Tuberculosis in Türkiye"* conducted through collaboration between Türkiye and the Netherlands in 2012, the individual prevalence of brucellosis in bovines was reported as 2.6%, while the herd prevalence was reported as 6.9% (Bartels et al., 2012). Prior to 2012, as part of the program aimed at fighting bovine brucellosis, the *B. abortus* S19 vaccine was administered subcutaneously to female calves aged between 4 and 8 months.

**Figure 1**. Number of animals compensated, and amount of compensation paid due to bovine brucellosis in Çankırı province (2011-2015.1*) (₺: Turkish lira) (MoAF)




*Based on the first six months data in 2015.

With the consent of the MoAF authority dated 31.12.2011, the *"Project for Control and Eradication of Brucella with Conjunctival Vaccine"* (Circular No: 2012/03) came into effect in 2012. Implemented within this framework, the project aimed to initially reduce the prevalence of brucella in bovine herds to less than 1%, followed by the complete eradication of the disease through the test and slaughter method. According to the project guidelines, starting from 2012, it was planned to administer the *B. abortus* S19 conjunctival vaccine to all female bovines twice, with an interval of 12 months. In subsequent years, the aim was to vaccinate adult female bovines that could not be vaccinated previously, as well as female calves aged between 3 and 6 months, thereby implementing a 10-year vaccination program.

In 2012, out of the 133,010 bovines in Çankırı province, 85,000 were enrolled in the vaccination program by MoAF. Throughout the province, vaccination was administered to 81,773 bovines, achieving a significantly high vaccination rate of 96.2% (Figure 2). In 2013, 95,750 out of 130,658 bovines were included in the vaccination program. Across the province, 79,031 bovines were vaccinated, resulting in a vaccination rate of 82.5%. In subsequent years, the program continued by vaccinating previously unvaccinated animals and female calves aged 3-6 months, based on vaccine availability. Furthermore, an informational brochure about brucellosis disease and the importance of vaccination was prepared and distributed to breeders and their organizations. The distribution of vaccines sent as part of the program throughout the province and the number of vaccinations administered by the teams were regularly monitored and reported to the MoAF, GDFC.

**Figure 2**. Çankırı province bovine brucellosis mass conjunctival vaccination studies (MoAF)

## DISCUSSION

The prevalence of bovine brucellosis is notably high in Mediterranean countries including Türkiye. In this country, MoAF has been engaged in efforts to control and eradicate this disease for many years. However, despite these efforts, the desired success has not yet been achieved, due to various reasons. Mass vaccination of bovine against brucellosis was identified as the most cost-effective measure compared to alternative methods. Therefore, it is recommended to utilize a vaccine strain that ensures robust immunity while also maintaining safety for both public and animal health.

In 2011, the burden of infection on bovine populations was reduced through the conditional slaughter and culling of bovine reservoirs of *B. abortus*, along with the disinfection of premises where the disease was present. As part of the project, Çankırı province was among the first provinces to implement a mass conjunctival vaccination program for bovines in 2012. By 2012 and 2013, a vaccination rate exceeding 82.5% resulted in robust herd immunity. Stakeholders were informed about these initiatives, and their support was garnered. As a result of these concerted efforts, the number of bovine brucellosis outbreaks and the subsequent compensation for affected animals decreased significantly in the subsequent years. While the percentage of outbreaks in Çankırı province, Türkiye, was 11.73% in 2011, this rate decreased to 2.85% in 2015 following the intervention efforts (Table). In addition, as shown in Figure 1, when compared to the 4.5-year total, the number of compensation payments (82.3%) and the amount of compensation paid (86%) were notably high in 2011. These rates were observed to decrease gradually each year in 2012, 2013, 2014, and 2015.1. As a result of this effective fight, in addition to reducing the risk of public and animal health, a significant amount of public resources have been saved and the socioeconomic loss of the sectors has been prevented. Verbal interviews with health institutions in the province indicated a decrease in the number of human cases as well.

After the vaccination of bovines, the cellular immune response is stimulated. The *B. abortus* S19

strain has been effectively used in the field to prevent infection for over eighty years (Dorneles et al., 2014; Dorneles et al., 2015). The availability of both conjunctival and subcutaneous forms of this vaccine, produced by both state and private sectors in Türkiye, offers significant advantages in terms of cost-effectiveness and logistical planning. It's essential to consider the long-term protection and cross-protection against *B. melitensis*. However, several challenges exist regarding the *B. abortus* RB51 strain. The exact nature of its mutations remains unknown, and its high cost, due to the absence of local production, presents obstacles for diagnosis and treatment. Additionally, its resistance to rifampicin and inability to be detected by routinely used serological tests further complicate matters, particularly in human cases (Schurig et al., 2002; Dorneles et al., 2015; Khurana et al., 2021). However, both vaccines can be administered to pregnant animals, although there is a risk of inducing abortion (de Oliveira et al., 2022).

WOAH recommends the use of 5-8x$10^{10}$ CFU dose of *B. abortus* S19 strain in female calves aged 3-6 months, along with one or two doses of 5x$10^{9}$ CFU administered via the conjunctival route in heifers and cows of all ages (de Oliveira et al., 2022). This recommendation suggests that the project has selected an optimal dose for infection prevention. The success of this research further confirms the validity of this dosage selection.

The number of diseases free premises and cattle must be sufficient to support this endeavour. Furthermore, vaccination of cattle in the vicinity of disease-free premises can significantly reduce the pathogen load, thereby minimizing the risk of disease transmission to these premises.

Brucellosis stands as a significant public health concern globally. Hence, it is imperative to foster collaborative efforts with all stakeholders within the framework of the One Health approach to reduce the prevalence of this disease. The MoAF and the Ministry of Health should both allocate funding to combat this ailment effectively. Raising awareness among relevant stakeholders, fostering a shared understanding, and maintaining open channels of communication are paramount for success in this endeavour (Bartels et al., 2012; Khurana et al., 2021). Disseminating information to breeders and their associations about the disease and vaccination through brochures has proven effective in increasing vaccination rates. Additionally, adopting practices such as burying discarded or deceased offspring and fetal membranes due to infection, rather than disposing of them into the environment, helps to reduce potential transmission sources. Moreover, individuals including breeders, caretakers, and workers who are educated about the zoonotic nature of the disease demonstrate increased vigilance against self-infection.

## CONCLUSION

As demonstrated by the example of Çankırı province, it is evident that controlling bovine brucellosis in Türkiye is feasible, albeit requiring substantial financing and a long-term commitment. Given the high prevalence of the disease in bovine herds, it is deemed achievable to attain eradication by initially reducing the prevalence below 1% through vaccination. Subsequently, a regional eradication strategy can be implemented gradually, utilizing the test-and-slaughter method. Efficient planning and uninterrupted provision of financing and free breeding stock are imperative at this stage. Adequate resources should be allocated to meet the breeding cattle needs of areas where the disease has been eradicated.

Activities should be meticulously planned and carried out in coordination as a state policy throughout the country. During the planning phase, various modelling approaches should be explored, prioritizing the selection of the most cost-effective option. When implementing test-and-slaughter

policies, public health considerations should take precedence, and the Meat and Milk Institution should be tasked with the public responsibility of slaughtering conditional animals. Given the advantages and risks, both subcutaneous and conjunctival formulations of the *B. abortus* S19 strain are recommended for use in Türkiye. To achieve optimal herd immunity, vaccination campaigns should aim for a coverage rate of 80% or higher. Effective communication with stakeholders and raising their awareness are vital for achieving set targets. Proper identification of animals, accurate record-keeping, and monitoring of their movements are essential components of successful disease management.

MoAF should promptly implement specialty training programs in Veterinary Medicine, while simultaneously increasing the number of specialized personnel in epidemiology and related fields, along with enhancing their professional rights. Support for research on vaccine development and disease diagnosis should be prioritized, with efforts directed towards updating diagnostic methods to align with international standards. Encouraging the training of specialized human resources is essential. Additionally, there should be an emphasis on expanding research and development capacity to create new vaccines and adjuvants that are safer, more effective, and suitable for field conditions.

In conclusion, since bovine brucellosis is a zoonosis and all stages of the fight against the disease require field expertise, a "One Health" approach should be adopted, and multidisciplinary studies should be carried out. It should not be forgotten that the protection of public health depends on the health of animals and food.

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**Ethical Statement**

Since retrospective data were used in this study, there was no need to obtain ethical permission.

**Author Contributions**

Research Design (CRediT 1) Author 1 (%60) – Author 2 (%40)
Data Collection (CRediT 2) Author 1 (%80) – Author 2 (%20)
Research - Data analysis - Validation (CRediT 3-4-6-11) Author 1 (%50) – Author 2 (%50)
Writing the Article (CRediT 12-13) Author 1 (%60) – Author 2 (%40)
Revision and Improvement of the Text (CRediT 14) Author 1 (%50) – Author 2 (%50)

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**Conflict of Interest**

Authors declare that there is no conflict of interest.

**Sustainable Development Goals (SDG)**

2 Zero hunger
3 Good health and well-being
8 Sustainable economic growth
12 Ensure sustainable consumption and production patterns

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