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Seroprevalence of Abortion-Related infectious diseases and associated risk factors among Brucellosis-free herds in Northern central Algeria

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Abstract

Background Cattle abortion, which may be caused by different infectious agents, harms milk and meat production, animal health, and ultimately rural economies. Despite the existence of a national control program for cattle brucellosis since 1995, abortion remains a major concern for cattle breeders in Algeria even among officially recognized cattle Brucellosis-free herds. The objective of this study is to investigate i: the abortion rate among officially recognized cattle Brucellosis-free herds, ii: the seroprevalence of some abortive infectious agents within cattle Brucellosis-free herds, iii: evaluate the abortion risk factors associated with the seropositivity of the investigated abortive agents among brucellosis cattle-free herds.

Results The present study reveals an abortion rate at the herd and the individual level of 47, 36% (20/38) and 29, 06% (50/172) respectively. Herd seroprevalences were as follows: neosporosis (31,57%), toxoplasmosis (28,94%); chlamydiosis (*Chlamidophila abortus*) (15,78%); Q fever (*Coxiella burnetii*) (47,36%); Bovine viral diarrhoea (BVD) (60,52%) and infectious bovine rinotracheitis (IBR) (42,10%). The risk factor analysis using a multivariable logistic regression model at the herd level showed that seropositivity to neosporosis (OR = 1, 11, CI: [0,85 – 1,19]); toxoplasmosis (OR = 1,95, CI: [1,22 – 2,84]); IBR (OR = 1,78, CI: [1,59 – 2,79]); BVD (OR = 1,65, CI: [1,86 – 3,43]); Q fever (OR = 1,51, CI: [1,42 – 2,53]) is a risk factor for abortion. Additionally, our findings reveal that the presence of co-infection is also a risk factor for abortion among Algerian Brucellosis-free herds. The risk for abortion at the herd level was 1,41 times higher for double and triple-infected herds and 1,65 times higher for quintuple-infected herds compared to negative. Moreover, managerial factors, such as hygiene practices on farms, the presence of primiparous cows, and mixed breeding livestock, were identified as additional risk factors for abortion.

Conclusion Based on these results, particular attention should be given to the studied abortive agents to strengthen the prevention and control plan. Furthermore, establishing some preventive measures such as quarantine and biosecurity could help reduce infections in dairy farms.

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Keywords Abortion, Cattle, Infectious agent, Seroprevalence, Risk factors, Algeria

Background

Abortion in cattle has several negative impacts on animal production, including damaging economic, reproductive, and animal health repercussions [1]. The significant economic impact is a combination result of a decrease in milk and meat production and an increase in veterinary and logistic expenses. Subsequently, the control of abortion in livestock and the prevention of this huge amount of economic loss are vital for cattle breeders [2].

Cattle production is one of the main sources of meat and milk production in Algeria that plays an important role in food security; the cattle population in Algeria stands at 2,2 million heads of which 70% is dairy cattle (30% imported cattle and 40% local improved cattle) representing an important food source to the inhabitants [3]. Algeria has significant potential in the field of cattle breeding, but it currently struggles with a substantial deficit in dairy and meat production. This deficit leads to a major import bill each year, which was reported by the Algerian Ministry of Commerce to be \$2.045 billion for milk powder and \$0.307 billion for meat in 2014 [4].

Cattle abortion can be attributed to various infectious agents, including viruses (bovine viral diarrhoea virus (BVDV), bovine herpes virus 1/4 (BoHV1/4), and the Schmallenberg virus); bacteria (*Brucella abortus*, *Chlamydomphila abortus*, *Coxiella burnetii*, *Leptospira interrogans* serovar *Hardjo*, *Salmonella enterica* serovar *Dublin*, *Campylobacter fetus* subsp. *Veneralis*, and *Listeria monocytogenes*); parasites (*Neospora caninum* and *Trichomonas fetus*), and more rarely fungi like *Aspergillus sp* [5, 6]. Additionally, non-infectious factors such as exposure to toxic substances, nutritional deficiencies, metabolic disorders, heat stress, and genetic abnormalities can also result in abortion [7].

Brucellosis is one of the most important abortive agents in dairy cattle, its occurrence in Algeria has been reported several times through different studies [8–10], despite the application of a national control program since 1995. On the other hand, abortion is still a major concern for cattle breeders even within herds officially declared Brucellosis free by the Algerian veterinary authority. Therefore, the objective of the present study is i: to assess the abortion rate, ii: investigate the seroprevalence of several abortive agents, and iii: evaluate the abortion risk factors associated with the seropositivity of the investigated abortive agents among brucellosis cattle-free herds.

Results

The present study reported a herd and individual abortion rate of 47,36% (20/38 herds) and 29,06% (50/172 cows) respectively.

Individual and herd prevalence

The results of the serological analysis of the sampled cows revealed that only 15 out of the 172 (8,72%) were negative for all the investigated infectious diseases. The remaining 157 cows, were positive to at least one disease; At the herd level, 10 out of 38 herds were reported negative to all the investigated infectious diseases.

The seroprevalence rates of the different studied diseases at the individual and herd level, including neosporosis; toxoplasmosis; chlamydiosis; Q fever; Bovine viral diarrhoea (BVD) and Infectious Bovine Rhinotracheitis (IBR) are described in Table 1.

Occurrence of abortion by co-infection status

The present study showed that 91,28% of the studied cows (157/172) were positive for at least one disease. Table 2 shows the serological status of cows according to their co-infectivity status.

At the herd level, the multivariate logistic regression analysis showed that the risk of abortion is 1,41 (CI 1,22–1,85, $p=0,041$) and 1,65 (CI 1,27–2,05, $p=0,044$) times higher in herds with double or triple infections and in herds with quintuple infections, respectively, compared to herds with no reported infections (Table 3).

Risk factors study

Table 4 shows the seropositivity occurrence of the studied diseases in cows according to their abortion status. Except for chlamydiosis, where the seropositivity in the aborted cows was 4%, the occurrence of the other diseases was markedly higher in the aborted cows.

At the herd level (Table 5), the multivariate logistic regression study demonstrated that the seropositivity of all the studied infectious agents, potentially present, a risk of abortion; the risk was 1,95 times higher (CI: 1,22–2,84; $p=0,041$), 1,65 times higher (CI: 1,86–3,43; $p=0,035$), 1,51 times higher (CI: 1,42–2,53; $p=0,045$), and 1,78 times higher (CI: 1,59–2,79; $p=0,044$) when associated with toxoplasmosis, BVD, Q fever, and IBR infections, respectively.

Managerial risk factors

The risk of abortion associated with managerial factors was performed by multivariate logistic regression study (Table 6), through which it appears that the risk of abortion is 1,22 times higher in mixed breeding of cattle and

Table 1 Description of the abortion and infection rate at individual and herd level

| | Neosporosis | Toxoplasmosis | Chlamydiosis | Q fever | BVD | IBR | Abortion |
|-----------------------------|---------------------|----------------------|---------------------|----------------------|------------------------|--------------------|----------------------|
| Positive cows | 26 | 26 | 7 | 64 | 128 | 68 | 50 |
| Negatives cows | 146 | 146 | 165 | 118 | 44 | 114 | 122 |
| Individual seropositivity % | 15,02 | 15,02 | 4,04 | 36,99 | 73,98 | 39,30 | 29,06 |
| Positive herds | 12 | 11 | 6 | 18 | 23 | 16 | 18 |
| Negative herds | 26 | 27 | 32 | 20 | 15 | 22 | 20 |
| Herd seroprevalence % | 31,57 | 28,94 | 15,78 | 47,36 | 60,52 | 42,10 | 47,36 |
| (CI) | (25,7–38,07) | (23,44–34,44) | (7,18–24,38) | (43,46–51,26) | (50,0.02–71,02) | (33,6–50,6) | (37,06–55,66) |

CI: Confidence Interval

Table 2 Description of the abortion rate by co-infection status at the individual level

| Abortion | Negative | Single | Double | Triple | Quadruple | Quintuple |
|----------------|-----------|------------|------------|------------|-----------|-----------|
| No ($n=122$) | 10 (8,2%) | 15 (12,3%) | 16 (13,1%) | 8 (6,6%) | 5 (4,1%) | 1 (0,8%) |
| Yes ($n=50$) | 5 (10%) | 33 (66%) | 48 (96%) | 22 (44%) | 9 (18%) | 0 (0%) |
| Total | 15 (8,7%) | 48 (27,9%) | 64 (37,2%) | 30 (17,4%) | 14 (8,1%) | 1 (0,6%) |

Table 3 Description of the abortion rate and associated co-infection status risk factors at herd level

| Abortion | Negative | Double/triple | Quintuple |
|----------------|------------|------------------|------------------|
| No ($n=20$) | 8 (44,4%) | 7 (38,9%) | 3 (16,7%) |
| Yes ($n=18$) | 2 (10%) | 11 (55%) | 7 (35%) |
| Total | 10 (26,3%) | 18 (47,4%) | 10 (26,3%) |
| OR | ref | 1,41 | 1,65 |
| 95% CI | / | 1,22–1,85 | 1,27–2,05 |
| P Value | / | 0,041* | 0,044* |

CI: Confidence Interval, OR: Odds Ratio, * $p < 0,05$

sheep (CI: 1,06–1,32; $p=0,043$), and 1,45 higher in farms with heifers (CI: 1,13–2,21; $p=0,046$) compared to reference herds.

In addition, the lack of control of newly introduced animals and the lack of hygiene in livestock building increases the risk of abortion by 1,89 (CI: 1,28–2,81; $p=0,022$) and 1,33 (CI: 1,19–1,69; $p=0,039$) times respectively compared to reference herds.

Discussion

Abortive Infectious agents represent a major health problem in cattle. They affect the reproductive and productive performances causing a major economic loss. However, the etiologic diagnosis remains challenging to determine because of the variability of the pathogen agents. Moreover, the aborted fetuses are subject to alteration and contaminations which further complicates the diagnosis [11].

Brucellosis is one of the major infectious agents known to cause abortion in cattle, however, after more than 20 years of the adoption of the national brucellosis control program (based on screening and slaughtering) in the country, the disease is now less frequent in controlled herds.

The objective of this study focuses on studying the prevalence of abortive infectious diseases (among brucellosis-free herds) and correlating their possible association with the occurrence of abortion by performing a multivariate logistic regression analysis.

Table 4 Seropositivity at the individual level

| Abortion | Neosporosis | Toxoplasmosis | BVD | Chlamydiosis | Q Fievre | IBR |
|----------------|-------------|---------------|-------------|--------------|------------|------------|
| No ($n=122$) | 11 (9%) | 6 (4,9%) | 78 (63,9%) | 5 (4,1%) | 19 (15,6%) | 18 (14,8%) |
| Yes ($n=50$) | 15 (30%) | 20 (40%) | 50 (100%) | 2 (4%) | 45 (90%) | 50 (100%) |
| Total | 26 (15, 1%) | 26 (15,1%) | 128 (74,4%) | 7 (4,1%) | 64 (37,2%) | 68 (39,5%) |

Table 5 Sero-infectious risk factor associated with the abortion at herd level

| Abortion | Neosporosis | Toxoplasmosis | BVD | Chlamydiosis | Fievre Q | IBR |
|----------------|------------------|------------------|------------------|------------------|------------------|------------------|
| No ($n=20$) | 3 (16,7%) | 3 (16,7%) | 9 (50%) | 1 (5,6%) | 8 (44,4%) | 8 (44,4%) |
| Yes ($n=18$) | 3 (15%) | 8 (40%) | 17 (85%) | 3 (15%) | 12 (60%) | 15 (75%) |
| Total | 6 (15,8%) | 11 (28,9%) | 26 (68,4%) | 4 (10,5%) | 20 (52,6%) | 23 (60,5%) |
| OR | 1,11 | 1,95 | 1,65 | 1,4 | 1,51 | 1,78 |
| 95% CI | 0,85–1,19 | 1,22–2,84 | 1,86–3,43 | 0,28–1,95 | 1,42–2,53 | 1,59–2,79 |
| P Value | 0,22 | 0,041* | 0,035* | 0,35 | 0,045* | 0,044* |

CI: Confidence Interval, OR: Odds Ratio, * $p < 0,05$

Table 6 Managerial risk factor at herd level

| Abortion | Mixed breeding (cattle + sheep) | Lack of control of newly introduced animal | Disinfection of livestock building; | Presence of primiparous cows |
|----------------|---------------------------------|--|-------------------------------------|------------------------------|
| No (n = 20) | 9 (50%) | 11 (61,1%) | 11 (61,1%) | 3 (16,7%) |
| Yes (n = 18) | 11 (55%) | 6 (30%) | 8 (40%) | 5 (25%) |
| Total | 20 (52,6%) | 17 (44,7%) | 19 (50%) | 8 (21,1%) |
| OR | 1,22 | 1,89 | 1,33 | 1,45 |
| 95% CI | 1,06 – 1,32 | 1,28 – 2,81 | 1,19 – 1,69 | 1,13 – 2,21 |
| P Value | 0,043* | 0,022* | 0,039* | 0,046* |
| | No = ref | Yes = ref | Yes = ref | No = ref |

CI: Confidence Interval, OR: Odds Ratio, * $p < 0,05$

Through this study, the rates of abortion at the herd and individual level are comparable to those reported by Kardjadj (2018) [12] and Hamza and Bouyoucef (2013) [13], who stated an abortion herd prevalence in Algeria of 41,33% and 40% respectively. At the North African level, Lucchese et al. (2016) [14] reported an abortion prevalence of 10,4% at an individual level and 48% at the herd level in Morocco, which suggests a similar epidemiological situation to Algeria.

Individual and herd prevalence

All the investigated diseases were detected in the studied herds at different prevalence rates, the most important rates were reported for viral diseases, namely BVD and IBR, followed by Q fever, then toxoplasmosis and neosporosis with similar rates, and at last chlamydiosis, which was reported in only 6 herds.

The epidemiological status of IBR and BVD is poorly known in Algeria. Only a few studies were reported on these diseases. Kaddour et al. (2019) [15] have reported a prevalence rate of IBR of about 14,16% at an individual level and 58,33% at a herd level, the study was performed in four districts in the northeast of Algiers, these results are within range to this study.

Regarding the BVD, our results are comparable to those stated by Guidoum et al. [16] who reported an individual prevalence of 59,9% (138 out of 234) with a 95% CI [49,0–70,7%] and a herd prevalence of 93,5% (29 out of 31) with a 95% CI [78,6–99,2%]. However, our results contrast with those reported by Derdour et al. (2017) [6], who noted a very low prevalence of IBR (boHV4/1) which was about 3,61% and 0,55% respectively, and of BVD (1,4%). This is probably due to horizontal transmission of pestivirus and herpes virus which can be favored in intensive production systems, although the prevalence is very low [17].

Our results suggest an endemic epidemiological situation of BVD in livestock in Algeria, especially since Feknous et al. (2018) study on small ruminants in the same area, reporting equally high prevalence rates (17,7%–68,2%) [18].

The Serological prevalence of Q fever reported in this study (36,99% and 47,36%) is higher than that stated by Djellata et al. 2019 [19] which was about 8,42% and 16,13% at individual and herd levels respectively in the northcentral of Algeria. On the other hand, our results were comparable to those obtained by Menadi et al. (2020) [20] who stated a herd prevalence of 45,56% (41/90) (95%CI 35,27–55,84%) in the same study area.

This study showed a lower seroprevalence of *chlamydiosis* than that reported by Djellata et al. 2019 [19] and Hireche et al. 2022 [21], who stated respectively a prevalence of 12,23% and 16,8% at the individual level and 29,84% and 30,9% at herd level. However, our results are higher than those observed in Algiers (Northcentral littoral), Jijel (North-Eastern littoral), and Tiaret (Western high plateaus) in which 0,83%, 1,08%, and 3,10% of dairy cows were respectively seropositive, as well as 19,04% of herds in Tiaret and 2,22% in Jijel [6, 22, 23].

Regarding neosporosis, our results are similar to those reported by Derdour et al. (2017) [6] and Djellata et al. (2019) [19] who stated respectively a prevalence of about 15% and 13,86%.

For toxoplasmosis prevalence, our results are comparable to several studies, the serological prevalence ranges from 13,86% [19] to 28,79% [24]. A meta-analysis study conducted by Ouchetati et al. (2021) [25] stated a prevalence of 20,04% in cattle.

Risk factor study

The risk factor analysis using multivariate logistic regression showed that (except *chlamydiosis*), the seropositive cows for the different studied diseases were statistically more subjected to abortion ($p < 0,05$). The risk of abortion associated with infectious disease has already been established through different studies at a global level [26, 27].

Regarding the co-infectivity status of herds, it has been observed that the risk of abortion was found to be statistically higher ($p < 0,05$) in multi-infected herds. Indeed, herds with multiple pathogen infections are more likely subjected to abortion due to a combination of factors

that weaken the animal's immune system and increase the risk of reproductive failure.

In this study, we observed that all aborted cows were positive for BVD. The immunosuppression associated with this viral infection might be due to changes in the number or function of immune cells in cows which can lead to an increase in the occurrence of disease and pathology of important severity when BVDV-infected cattle are co-infected with other pathogens [28]. Moreover, multiple infections can lead to increased inflammation in the cow's reproductive system making them more prone to abortion [29].

Abortion in multi-infected cows can also be linked to nutritional and hormonal factors; indeed, multi-infected cows may experience nutritional stress which can weaken the cow's immune system and as a result, their ability to maintain the pregnancy [30].

Besides, infections can interfere with the hormonal signals that regulate the cow's reproductive system, leading to hormonal imbalances that can increase the risk of abortion [31].

Managerial factors have also been recognized as contributors to the increase in the risk of abortion especially in mixed breeding farms between cattle and sheep, where different pathogens could be hosted by animals facilitating the transmission of infections. Moreover, heifers are more subjected to abortion, probably because their reproductive system is not fully developed and their less immunity which makes them more susceptible to infections.

Conclusion

The results of the present study showcase a worrisome presence of abortive infectious diseases that need to be thoroughly investigated. Furthermore, the Algerian control program for cows focuses only on brucellosis neglecting the other reproductive diseases.

Based on the high seroprevalence reported in this study, and the risk posed by infectious agents and managerial factors in the occurrence of abortion, particular attention should be directed towards these infectious pathologies to strengthen the control plan by establishing good biosecurity practices, regarding preventive measures such as quarantine procedure and mass vaccination, which are essential for controlling the spread of diseases and reducing the risk of abortion in cattle.

Methods

The study was conducted during the period between 2018 and 2021, as a part of PRFU project N° D00L01EP160320180001 and was approved by the Scientific Council of Laboratory Research of the National High Veterinary School of Algiers.

All methods used in the present study were carried out per relevant guidelines and regulations. Veterinarians handling animals performed best practices per the WOAH's (formerly OIE) ethical guidelines and animal welfare regulations.

The scientific and ethical council of the laboratory research of the National High Veterinary School of Algiers determined that this study does not include any intervention with human subjects or any access to identifiable private information, and therefore does not require IRB (Institutional Review Board) review.

Study area

Algeria is a North African / Mediterranean country, located between latitudes 19 and 37° N and longitudes 9° W and 12° E. It is considered the largest country in Africa. Governmentally, it is organized into 58 districts. The present study was performed in the northern (coastal) central part of the country (35.3–36.8° N and 1–4.7° E).

Study design and sample collection

A cross-sectional study with a one-stage selection design as described by Hosmer and Lemeshow (2013) [32] was carried out between January and December 2019 to investigate abortion history in Algerian Brucellosis-free cattle herds between 2018 and 2019.

The herd size ($n=38$ herds) was determined at a 95% confidence level using an expected prevalence of 40% [13] and an absolute precision of 14%. All the cattle found within each herd were sampled. Since clinical abortion cannot be detected by farmers before the third month of gestation, only reports of abortions observed after this period were considered in this study.

The study herds include 172 cows (brucellosis-free) with a herd size ranging between 3 and 7 cows per herd. All cattle within the selected herds were sampled and investigated. All studied herds and cattle were only vaccinated against rabies.

Cattle owners (farmers) participating in the study were notified and informed about the objectives of this research investigation and their consent regarding cattle data/blood collection was obtained. All information about the abortion history, abortion frequency, and management risk factor attributes were collected for the risk factors analysis.

The blood samples were taken by the veterinarians from the caudal vein of every cow using a Vacutainer tube. The samples were obtained within a timeframe of two months after abortion, to reduce the likelihood of detecting a very low level of antibodies. Following collection, the samples were transported to the laboratory in a coolbox, maintained at a temperature of +4 °C, and subsequently centrifuged for 5 min at 3,000 rpm. Afterward,

the sera were preserved at -20°C until serological testing was performed.

Laboratory testing

The following ELISA kits (IDVET, Montpellier, France) were used according to the manufacturer's instructions to detect the presence of antibodies against the studied abortive agents.

ID Screen® IBR gE Competition for Bovine Herpes virus (BHV-1) agent of IBR (Infectious Bovine Rhinotracheitis),

ID Screen® Toxoplasmosis Indirect Multi-species for *Toxoplasma gondii* (Toxoplasmosis),

ID Screen® *Neospora caninum* Indirect for *Neospora caninum* (neosporosis),

ID Screen® Q Fever Indirect Multi-species for *Coxiella burnetii* (Q fever),

ID Screen® *Chlamydomphila abortus* Indirect Multi-species for Chlamydiosis.

ID Screen® BVD p80 antibody competition for Bovine Viral Diarrhoea (BVD).

Statistical analysis

An exploratory data analysis (EDA) was performed for the selection of variables with $P \leq 0.2$ by chi-square test or Fisher's exact test (univariable analysis). Afterward, the variables that passed this cut-off were exposed to multi-variable logistic regression.

The fit of the final model was verified using Hosmer and Lemeshow test [32], and collinearity between independent variables was verified by a correlation analysis. For variables with strong collinearity (correlation coefficient > 0.9), one of the two variables was excluded from the multiple analysis according to the biological plausibility [32]. Confounding was assessed by monitoring the changes in the model parameters when adding new variables as described by Dohoo et al. (1996) [33].

The Statistical analysis/calculations were performed using R (4.1.0) and RStudio (1.4.1717) for Windows. All tests were 2-sided and $p < 0.05$ was considered statistically significant.

Abbreviations

| | |
|-------|-------------------------------------|
| BoHV | Bovine Herpes virus |
| BVD | Bovine Viral Diarrhoea |
| CI | Confidence interval |
| EDA | Exploratory data analysis |
| ELISA | Enzyme-linked immunosorbent assay |
| IBR | Infectious bovine rhinotracheitis |
| OIE | Office international des epizooties |
| OR | Odds ratio |
| WOAH | World Organization Animal Health |

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Author contributions

FY and MK designed the study and contributed equally under the supervision of MHB. FY and SM performed the laboratory analyses. MK performed the data analyses. FY and MK wrote the manuscript. MS and MHB were involved in some sections of the draft manuscript and revised it critically. All co-authors read and approved the final manuscript.

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Data availability

The datasets collected/generated during and/or analyzed during the current study are available on reasonable request to the corresponding author.

Declarations

Ethics approval and consent to participate

The study was conducted between 2018 and 2021, as a part of PRFU project N° D00L01EP160320180001, and was approved by the Scientific Ethical Council of Laboratory Research of the National High Veterinary School of Algiers. Cattle owners (farmers) participating in the study were notified and informed about the objectives of this research investigation and their informed consent regarding cattle data/blood collection was obtained. All methods used in the present study were carried out per relevant guidelines and regulations. Veterinarians handling animals perform best practices according to the WOA's (ex OIE) ethical guidelines and animal welfare regulations.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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