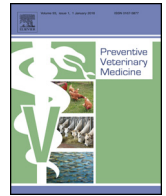




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Abortion and various associated risk factors in small ruminants in Algeria

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ABSTRACT

Identification of the causes of abortion among the huge population of small ruminants in Algeria (≈ 31 millions heads), is an important task for the control of livestock productivity and viability scourges to the small ruminants industry. Optimal production and utilization is constrained by a number of factors: disease, poor feeding and low management skills. Therefore, in the present study the prevalence of abortion in Algerian small ruminant's flocks was estimated and its possible association was correlated with infectious (PPR, BT and Brucellosis seropositivity) and managerial (flock size, grazing system, type of farming, and contact with other flocks) risk factors. The present study showed an overall flock prevalence of small ruminant's abortion as 75.33% (113/150) [95% CI 71.72–78.94%]. The risk factor analysis using multivariable logistic regression recognized the north-western and the steppe region as well as PPR positivity as a risk factor for abortion in Algerian small ruminant's flocks. The odds of flock abortion was 11.47 [95% CI 2.39–54.88; $P=0.002$] and 10.31 [95% CI 1.28–82.88; $P=0.028$] times higher in north-western and steppe regions respectively compared to other region. Also the presence of PPRV infection in small ruminant flocks amplified the odds by 6 times [95% CI 2.221–17.427; $P=0.001$]. Surprisingly, the univariate analysis for the other risk factors associated with abortions in Algerian small ruminant flocks indicated no statistically significant links with bluetongue ($P=1.000$) and brucellosis seropositivity ($P=0.334$). Flock size ($P=0.574$), type of farming ($P=0.443$), grazing system ($P=0.117$) and contact with other flocks ($P=0.245$) was also not statistically significant. Our results revealed that abortion in small ruminants is a challenge to farmers and PPR was chiefly linked to it. Therefore an effective vaccination and control programme is advocated for small ruminants in Algeria.

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1. Introduction

Small ruminant production is one of the main sources of meat production (≈ 31 millions heads) in Algeria that plays a vital role in food security. The small ruminants industry has the ability to improve the living standards of farmers and households, as well as increase animal protein for Algerians and consequently alleviating poverty. The small ruminant population in Algeria stands as 27 millions sheep and 4 millions goats respectively, where 65% of the total populations are females and 35% males. However, both sheep and goats are reared under traditional extensive system in Algeria,

intensive husbandry systems has recently been introduced in the country (MADR, 2014).

In spite of the population advantage of small ruminants, diseases and poor herd-health management practices poses a significant challenge to optimal and efficient management and profitable small ruminants' production in developing world such as Algeria. However, the viability of sheep and goat farming depends largely on their reproductive performance which is invariably regulated by genetic and environmental factors (Mellado et al., 2006). Additionally, abortion represents the most dangerous livestock productivity and viability scourges and public health concerns posed by some of the zoonotic microorganism (Van Engelen et al., 2014; Benkirane et al., 2015).

Abortion of food producing animals have a negative impact on livestock production, animal health and ultimately rural economies since most small ruminants are kept by the rural poor as a means of

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alleviating poverty (Diallo, 2006). However, the farming system and communal grazing practiced enables infectious organism to spread very fast and therefore, there is the need for an improved diagnostic capacity, appropriate and adequate control strategies and periodic monitoring for healthy livestock and public health safety (Van Engelen et al., 2014). There are several potential factors underlying the causes of abortion which can be broadly categorized into infectious and non infectious (Entrican, 2009).

Infectious agents are the most plausible causes of abortion in sheep and goats as compared to non-infectious agents and are mostly zoonotic. The main infectious causes of abortion in sheep and goats are *Chlamydia abortus*, *Campylobacter* spp., *Toxoplasma gondii*, *Listeria* spp., *Yersinia pseudotuberculosis*, *Coxiella burnetii*, *Brucella melitensis*, and *Bluetongue virus* (BTV) (Entrican, 2009; Van den Brom et al., 2012; Lafi et al., 2013; Ababneh et al., 2014).

North Africa and the Middle East has been described as endemic areas for Brucellosis with characterized high incidences of human cases (Rubach et al., 2013). In North Africa, this scenario is supported by environmental factors, communal grazing mostly by the poor farmers (Spickler and Roth, 2008). Exposure to these pathogens can predispose an animal to abortion and severe public health implications (Holler, 2012; Benkirane et al., 2015).

Infection from early embryonic term to the end of the gestation period plays a critical role in the viability of the fetus. These diseases cause abortion, fetal loss and congenital abnormalities in lambs and kids (Radostits et al., 2007; Van Engelen et al., 2014). Recently, Peste des Petites Ruminants Virus (PPRV) infection of pregnant sheep and goats has been implicated in abortion in Turkey (Güler et al., 2014). Subsequently, Kardjadj et al. (2015a) described the first occurrence of the disease in both sheep and goats in Ghardaïa district, the isolated strain belongs to lineage IV of PPRV and was genetically closely related to the strain identified in neighboring Morocco and Tunisia. This infectious and highly contagious agent spreads easily among small ruminants in a system with low management practices. Furthermore, the fact that both sheep and goats are not vaccinated against PPRV makes this disease a serious threat to the livelihood of Algerian farmers.

In 2012, Hamza and Bouyoucef (2013) reported that 90% of Algerian farmers had observed abortions in their heads. However, the causes of these abortions remained undiagnosed. In order to protect and sustain the Algerian small ruminant industry and equally boost productivity, it is therefore critical to have adequate knowledge about the possible abortion causes. The present study attempted to investigate and estimate the prevalence of abortion in Algerian small ruminant's flocks during a national survey and correlate its possible association with infectious agents (PPR, BT and Brucellosis seropositivity) and managerial (flock size, grazing system, type of farming, and contact with other flocks) risk factors.

2. Materials and methods

2.1. Study area

Algeria is located between latitudes 19° and 37°N and longitudes 9°W and 12°E. It is the largest country in Africa. It has a long coastline at the Mediterranean Sea (1600 km); Most of the coastal area (northern region) is hilly, sometimes even mountainous. South of the northern region is a steppe; farther south, there is the Sahara desert. Administratively, Algeria is divided into 48 districts (wilayas) but for the sake of this study and according to the geographical and farm management specificity, the country is delineated into five regions, with each region containing 7 to 12 districts; north-central (35.3°–36.8°N and 1°E–4.7°E), north-western (35°–36.3°N and 2°W–1°E), north-eastern (35.3°–37°N and 4.7°E–8.5°E), steppe (33°–35.3°N and 2°W–8.5°E) and Sahara region (19°–33°N and 8.8°W–12°E).

2.2. Study design and sample collection

Small ruminant owners participating in the study were informed about the purpose of the study and their verbal consent was obtained. The number of flocks to be sampled from each region was proportional to the percentage of small ruminants in that region. Flocks were selected from each region using random numbers generated by an electronic calculator.

A cross-sectional study with a two-stage selection design as described by Toma et al. (2009) was carried out for 6 months across the country between January and June 2014. The simple size ($n = 150$ flocks) and the number of animals to be sampled within each flock ($m = 15$ animals) was determined at a 95% confidence level using; an expected prevalence for bluetongue at 15% and brucellosis at 5%, respectively (Madani et al., 2011; Kardjadj, personal communication), with an absolute precision of 4% and 2.5%, an estimated within-class coefficient of $P = 0.4$ and 0.6 and an inflation coefficient of 7 and 10.

From the 150 flock, a total of 2421 blood samples were collected (1,932 sheep and 489 goats) by jugular venous puncture in 5 ml sterile vacutainer tubes using venoject needles (Venject, UK). Blood samples were transported on ice to Institut National de Medecine Vétérinaire (INMV), Algiers, Algeria for analysis. The samples were centrifuged at 3000 rpm for 5 min and separated into a sterile tube and stored at -20°C until tested.

2.3. Laboratory analysis

The 150 selected flocks were screened for antibodies to PPR (Kardjadj et al., 2015b), using a competitive ELISA (c-ELISA) according to manufacturer's instructions (ID Screen® PPR Competition, ID vet, Montpellier, France). This diagnostic kit detects antibodies against the nucleoprotein of PPRV with sensitivity and specificity of 94.5% and 99.4%, respectively (Libeau et al., 1995).

Similarly, a competitive enzyme-linked immune-sorbent assay (c-ELISA) was performed for BTV antibodies using a commercially c-ELISA Kit (Veterinary Medical Research and Development Laboratory, USDA Pullman, WA, USA). The sera were screened for IgG anti-VP7-BTV antibodies as described by the manufacturer's specifications. The test sensitivity and specificity were 100% and 99%, respectively according to the manufacturer's specification.

The assay was performed in a 96-well antigen coated microplates. The incubations were performed for 15 min at room temperature ($21 \pm 2^{\circ}\text{C}$). The plates were washed three times with the provided washing buffer. Briefly, aliquots of 25 μl test sera as well as positive and negative controls sera were transferred undiluted to the BTV antigen coated plates. After incubation, the plates were washed, and 25 μl of antibody-peroxidase conjugate were added to each well. The plate was then incubated at 15 min at room temperature. The plates were then washed and 50 μl the substrate was added to each well. The reaction was stopped using 50 μl of the stopping solution. The results were read using ELISA reader set at 630 nm. A diagnosis was made when the test samples produced an optical density <50% of the mean of the negative controls. The test samples were considered negative if the optical density $\geq 50\%$ of the mean of the negative controls.

Additionally, all serum samples were screened using Rose Bengal Plat Test (RBPT) according to the procedures (Lilidale®). Briefly, 75 μl of serum was mixed with 25 μl antigen, on the plate and shaken. After 4 min of gentle shaking, any visible agglutination was considered as positive.

2.4. Data collection and statistical analysis

A questionnaire was administered to small ruminant owners (farmers) in all selected flocks by personal interview with 100%

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