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Serological review of etiological specialists related with fetus removal in two Algerian dairy steers reproducing ranches

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Abstract

In Algeria, there is a lack of information about the frequency and etiology of abortions in breeding cattle. In order to improve this situation, we sent a questionnaire to veterinary practitioners, and followed it up with a serological survey on two dairy farms (A and B) that had reported abortions. The serological survey was undertaken to detect the main abortive agents with particular emphasis on *Neospora caninum*, a parasite presently classified as one of the most important abortive entities globally, but still ignored in our country. The results of the questionnaire showed that when an abortion occurs, no particular measure is taken by practitioners as a diagnosis of the causative agent is very difficult to achieve due to lack of resources. The serological survey showed an average positive rate of 0, 32.8, 37.5 and 50% respectively for brucellosis, neosporosis, candidiasis and bovine rhinotracheitis in both herds and a rate of 3.2, 9.6 and 29% respectively, for chlamydiasis, salmonellosis and Q fever in farm (B) alone. This preliminary study, therefore, indicates for the first time that neosporosis exists in some Algerian farms and may contribute, either individually or in association with other agents to abortions in breeding cattle.

Keywords: Abortion, Algeria, breeding cattle, serology, *Neospora caninum*, livestock diseases.

INTRODUCTION

Abortion of infectious origin is considered a significant pathology given the considerable economic losses due to loss of production income (e.g. loss of calf and milk), on the one hand, and loss of breeding stock due to compulsory slaughters imposed in cases of suspected brucellosis on the other. For example according to Je-In and Ill-Hwa (2007), the cost of abortion in Korea is estimated at $2,333/cow ; while, Reitt et al. (2007) report that with an abortion rate of 2 to 4% annually in Switzerland, the loss is estimated as being between 22.4 and 44.8 million Swiss francs (1 CHF = 1USD). In Algeria, the amount of compensation for cattle and goats slaughtered because of brucellosis between 2002 and 2004 was estimated at 83 million Algerian dinars (1 DZD = 0.139 USD).

Infectious abortion is caused by many bacteria, viruses, protozoa and fungi, and the diagnosis of the exact cause is not easy in all cases. According to Anderson (2007) and Kradel (1978), the etiology is identified in less than half of the cases submitted to laboratories. Serology does not allow a precise diagnosis although it does determine whether there has been exposure to an abortive agent or not.

In Algeria, little is known about the epidemiology of infectious abortions. The lack of data and information relative to the frequency of abortions and to the probable origins in our farms is due to the fact that reporting is not obligatory and laboratory diagnosis is difficult due to lacks of funds and suitable diagnostic facilities. The limited data that is available is out-dated and focused primarily on brucellosis, or only on the identification of a single infectious agent. Examples of such studies include bovine listeriosis (Bellouni and Rahal, 1992), BHV-1 (Benazzouz, 1981; Achour and Moussa, 1996) and *Coxiella burnetti* by Boudjemaa (1987).

Neosporosis, an abortive pathology of a relatively recent discovery, has been identified in many countries (for review see Dubey et al., 2007). To date, no study has

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been published on the presence of neosporosis in Algeria and the prevalence of the disease still remains unknown.

The aim of the present study was, therefore, to firstly determine via a questionaire the epidemiological, clinical and etiological situation of abortions in dairy cattle and the action adopted by farmers and veterinarians when confronted by these cases. Secondly, we looked for serological traces of a number of abortive agents in two dairy farms that had recorded sporadic cases of abortions with particular emphasis on anti-*Neospora caninum* antibodies.

MATERIALS AND METHODS

Questionnaire

A questionnaire (copy available on request) was sent to 250 veterinarian practitioners in the central region of the country.

Site of study and animals

Our study was undertaken in two, semi-intensive dairy farms with a total of 237 cattle, of Montbeliarde and Prim'Holstein breeds that had suffered sporadic abortions estimated at between 6 and 8% per year. The first farm (A) was located in the region of Tipaza and the second (B) in the region of Blida. Both farms are certified by the Algerian veterinary services and are controlled biannually. They have been declared free of both brucellosis and tuberculosis. The number of animal samples was established based on the study budget and the cost of the diagnosis.

Samples

A total of 64 blood samples representing more than 20% of the cattle in each farm (33/87 head in farm "A" and 31/150 head in farm "B") were collected from the coccygeal vein in 10 ml vacuum blood tubes. Sera were separated by centrifugation and stored at -20°C until testing.

Serological methods

Serological analysis of brucellosis was conducted in the laboratory of Hygienic and Sanitary Quality of Milk (University Saad Dahleb-Blida) while for the other abortive agents; sera were analyzed in the laboratory of Reproductive Pathologies, Nantes National Veterinary School, France.

The Rose Bengal Test was used to detect *Brucella* specific agglutinins (*Brucella melitensis, Brucella abortus bovis* and *Brucella suis*) as described by Alton et al (1975) . This test uses a *Brucella abortus* suspension (strain S99) in lactate buffer 1 mol/L (SPINREACT, Spain) . The presence of agglutination indicates an anti- *Brucella* antibody concentration equal or greater than 25 IU/ml, with values of 93% sensitivity and 100% specificity.

For BHV-1, an ELISA [Laboratoire service international (LSI) kit-Lissieu- France] was used. The sensitivity and specificity values of the LSI test were 99% and 99.5% respectively. According to the manufacturer's instructions serum samples with an S/P ratio equal or greater than 1.00 were classified as positive.

Anti-*Neospora caninum* antibodies were detected by ELISA (HerdChek* kit, IDEXX, USA) which has a reported sensitivity of 98.6% and a specificity of 98.9%. According to the manufacturer's instructions the presence of *Neospora* antibodies is determined by an S/P ratio greater than or equal to 0.50.

Candida guillermondi antibodies were detected by ELISA. The antigen used was KH6m, which is a culture supernatant of *C. guillermondi* manufactured in the laboratory of Reproductive Pathologies, ENV, Nantes, France. A serum is considered positive if its optical density is superior or equal to 0.20 (i.e. twice the optical density of the negative serum).

The detection of anti-*Salmonella* antibodies was done only for farm "B". It was performed with a slow sero-agglutination test (18 hours at 37°C) using a coloured suspension of the "H" antigen of *Salmonella abortus ovis*. This test is manufactured locally and has been calibrated with a similar test developed by the Sophia Antipolis Laboratory, AFSSA, France. An agglutination of 50% at a dilution of 1:160 was considered positive.

Chlamydia psittaci antibodies were detected in farm "B" only using the complement fixation test (CFT) according to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (2005). The antigen was provided by Eurobio Laboratories, Paris, France. Sera giving a titre of 4+ at a dilution of 1:32 or greater were assumed to be positive.

A micro titer complement fixation test for *Coxiella* antibodies was performed only for farm "B" according to the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (2005). It was performed using a *C. burnetii* antigen provided by Symbiotics Europe, Lyon, France. It was a mixture of Henzerling and Nine Mile strains in Phase II produced in embryonated eggs. The presence of 50% haemolysis starting from a 1:10 dilution of sera was considered positive.

Statistical analysis

Statistical analyses were conducted using STATISTICA 6 Base (StatSoft, Inc., 2001). Seropositivity was calculated by dividing the number of serological positive animals by the total of sampled animals.

RESULTS

Questionnaire

Analysis of the responses to the questionnaire revealed that 85.7% of the practitioners encountered abortions sporadically while 25.7% of them encountered them on a monthly basis. Sixty per cent of the veterinarians thought that the probable cause for the abortions was brucellosis (even without laboratory confirmation) while only 2.8% thought that neosporosis might be the causative agent.
Of the practitioners 58.5% said that they were called by the farmer only when the abortion was followed by retained placenta. Veterinarians (47.5%) answered that they do not report abortions or take any particular measures to diagnose cause.

Serology

The results from our analysis showed that the two farms were free of brucellosis. For the other agents, investigated different seropositivities were recorded as shown in Table 1.

Table 1. Distribution of the seropositivities for abortion associated agents.

	Brucella spp	*BHV-1*		*Neospora caninum*		*Candida guillermondii*		*Salmonella abortus ovis*		*Chlamydia psittaci*		*Coxiella burnetti*	
	%	%	CI95%	%	CI95%	%	CI95%	%	CI95%	%	CI95%	%	CI95%
Farm A (n = 33)	0	66.6	(50.6 - 82.6)	30.3	(14.9-45.7)	39.3	(22.7-55.9)	/		/		/	
Farm B (n = 31)	0	32.2	(16 - 48.4)	35.4	(18.8-52)	35.4	(18.8-52)	3,2	(0-9.2)	9.6	(0-9.8)	29	(13.2-44)
Average rate	0	50	(37.8 - 62.2)	32.8	(21.5- 44.1)	37.5	(25.8-49.2)	/		/		/	

Mixed infection

A serological association of two or more agents was noticed in 24 of the sera tested: 19 sera were seropositive for two agents simultaneously one of which was *N. caninum*; 3 serums were seropositive for three agents simultaneously one of which was *N. caninum*; one serum was seropositive for *Candida*, BHV-1 and Salmonella simultaneously while one was seropositive for four agents simultaneously one of which was *N. caninum*.

DISCUSSION

The results from our questionnaire showed that the number of abortion cases in Algeria is significant. However, farmers do not consider them to be of great importance and veterinarians attribute most of the cases to brucellosis without looking for the real etiology. Similar results to ours have been reported by Lounes and Bouyoucef (2009) who showed that 27% of practitioners encounter abortions once a month with 78% of them considering brucellosis as the primary cause.

Of interest in this preliminary study is that while both farms were seronegative for brucellosis seropositivities for other abortion associated agents were detected.

The average neosporosis seropositivity of 32.8% confirms the existence of *N. caninum* in Algerian breeding cattle. There were no statistically significant differences (P = 0.665) in the seropositivity between the two farms. This rate is close to seroprevalences reported in other countries; e.g. 25.5% has been reported by Haddad et al., (2005) in Canada, 34.8% by Guinot (2005) in France, 35.4% by Lopez-Gatius et al., (2004) in Spain and 49% by Thompson et al. (2001) in Portugal.

One hypothesis for the existence of neosporosis in Algeria could be that this parasite has been imported by dairy cattle coming from countries such as France, United Kingdom and Canada countries which show high seroprevalence of neosporosis (Amellal, 1995).

A second hypothesis could be that the neosporosis was spread due to the presence of dogs, chickens, ducks and pigeons on the farms studied. The role that these animals can play in the transmission of the disease should not be neglected (Losson and Bourdoiseau, 2000). For example, it has been reported in the literature that chickens andpigeons may be an intermediate host for the neosposoris parasites (Costa et al., 2008; Mineo et al., 2009).

The data on the seropositivity of other abortion associated agents on the two farms is of value. BHV-1 is found worldwide with seroprevalence values of 42.5%, 85.8% and 50.9% reported in England, Croatia and India respectively (Woodbine et al., 2009; Biuk-Rudan et al., 1999; Renukaradhya et al., 1996). A seroprevalence of 20.5 and 45.6% in central and eastern regions of Algeria have previously been reported (Achour and Moussa, 1996; Benazzouz, 1981).The values of 50% reported in this study are therefore in agreement with other countries.

Seropositivity for candidiasis also showed a high rate in both farms at an average of 37.5%. Again, mycotics are reported worldwide. In some regions it is the major cause of abortion (McClausland et al., 1987).

Chlamydiosis seropositivity was recorded as 3.2% in farm (B). This is relatively low compared to the seropositivity of the other abortive agents. However these values are in agreement with other studies worldwide. For example, Akakpo et al. (1994) reported2.3% prevalence in Togo; 11.2% was reported by Ghirotti et al. (1991) in Zambia and 7.1% was observed in Czechoslovakia by Lisak et al. (1989).

A seropositivity of 9.6% for salmonellosis was shown for farm (B). In the United States, abortions attributed to Salmonella species are not common. However, in the United Kingdom, Salmonella abortion is an important cause of both enzootic and epizootic abortion (Hinton, 1977).

Cattle are considered in some countries as the major reservoir of *Coxiella burnetii,* in particular in Japan (To et al., 1998) and Canada (Lang, 1989). In Algeria, a previous serological study for Q fever was carried out in various areas of the east of the country, revealing rates which varied from

2 - 5.92% (Boudjemmaa, 1987). Therefore, the seropositivity in 29% of the cattle tested in farm (B) for Q fever is relatively high.

In the present study, there was a serological association between neosporosis and two or more other abortive agents. Similar finding were reported by Akakpo et al., (1994) where brucella, chlamydia and coxiella were associated with the same abortion. Also, Thurmond et al., (1997) reported involvement of *N. caninum* together with *Leptospira* spp. in abortion while Jeffrey and Hogg (1988) showed an association between *N. caninum* and *Campylobacter foetus.*

It is important to point out that in mixed cases it is extremely difficult to establish the actual agent responsible for the abortion. Indeed, several authors claim that the responsible infectious agent is only diagnosed in 30 - 50% of cases (Radostis et al., 1994; Corbellini et al., 2006; Kim et al., 2002).

Another key issue to consider from this study is that serology alone does not determine the causative agent as the tests cannot distinguish between vaccinated or naturally infected animals. Likewise, serology will not determine whether and animal has been recently or historically exposed. Certainly increasing the number of samples taken within a herd is more informative. Also, taking samples at different time points will provide important information particularly if the animal has been exposed to the infectious agent between sampling periods.

Conclusion

This study is limited in size and scope given that only two farms were sampled. Nevertheless, the data obtained provides a good starting point for future and larger studies to determine the causative agents in abortion among breeding cattle. In these future experiments confirmation by isolation of the infectious agent will be essential in order to reach solid conclusions. The data obtained also provides a list of alternative causative agents, particularly neosporosis, to the veterinary practitioners in Algeria who up to now appear to associate abortions with brucellosis alone.

REFERENCES

Achour HA, Moussa A (1996). Serological and virological studies on the infectious bovine rhinotracheitis in Algeria.1: Zentralbl Veterinarmed B Jun; 43(4): 251-6.

Akakpo AJ, Teou KL, Kponmassi T, Zeller HG (1994). Epidémiologie des affections abortives au Togo: Enquête sérologique sur la brucellose, la chlamydiose, la fièvre Q et la fièvre de la vallée de Rift. Biotechnologies du diagnostic et de la prévention des maladies animales p. 125-137.

Alton GG, Maw J, Rogerson BA, McPherson GG (1975). The serological diagnosis of bovine brucellosis: An evaluation of the complement fixation, serum agglutination and rose Bengal tests. Aust. Vet. J. 51: 57-63.

Amellal R (1995). La filière lait en Algérie: entre l'objectif de la sécurité alimentaire et la réalité de la dépendance. CIHEAM-Options Méditerranéennes. Sér.B/ n° 14.

Anderson ML (2007). Infectious causes of bovine abortion during mid-to late-gestation Theriogenol. 68: 474–486.

Bellouni R, Rahal K (1992). Prospective study of *listeria* in humans and animals. Arch. Inst. Pasteur Alger. 58: 125-44.

Benazzouz M (1981). Contribution à l'étude de la rhinotrachéite infectieuse en Algérie, sondage sérologique. Mémoire en vue de l'obtention d'un diplôme de docteur vétérinaire.

Biuk-Rudan N, Cvetníc S, Madic J, Rudan D (1999). Prevalence of antibodies to IBR and BVD viruses in dairy cows with reproductive disorders. Theriogenol. Apr. 1-51(5): 875-81.

Boudjemaa N (1987). Etude de la fièvre Q chez les bovins:enquête sérologique dans les wilayas de Annaba et d'El Tarf. Mémoire rédigé en vue de l'obtention du diplôme de docteur vétérinaire.

Corbellini LG, Pescador CA, Frantz F, Wunder E, Steffen D, Smith DR and Driemeier D (2006). Diagnostic survey of bovine abortion with special reference to *Neospora caninum* infection: Importance, repeated abortion and concurrent infection in aborted foetuses in Southern Brazil. The Vet. J. 172(1): 114-120.

Costa KS, Santos SL, Uzêda RS, Pinheiro AM, Almeida MA, Ara'jo *FR, McAllister MM, Gondim LF (2008). Chickens (Gallus domesticus) are natural intermediate hosts of *Neospora caninum*, Int. J. Parasitol. 38: 157-159.

Dubey JP, Schares G, Ortega-Mora LM (2007). Epidemiol and Control of Neosporosis and *Neospora caninum* Clinical Microbiol. Rewiews, 20(2): 323-367, doi:10.1128/CMR.00031-06.

Ghirotti M, Semproni G, De meneghi D, Mungaba FN, Nannini D, Calzetta G, Paganico G (1991). Sero-prevalences of selected cattle diseases in the Kafue flats of Zambia. Vet. Res. Commun 15(1): 25-36.

Guinot PAG (2005). Etude séroépidémiologique de la néosporose bovine dans 42 élevages des Pyrénées-Atlantiques thèse pour obtenir le grade de docteur vétérinaire. Ecole nationale vétérinaire de Toulouse.

Haddad, JPA, Dohoo IR, VanLeewen JA (2005). A review of Neospora caninum in dairy and beef cattle-a Canadian perspective, Can. Vet. J. 46: 230–243.

Hinton M (1977). The diagnosis of salmonella abortion in cattle with particular reference to *Salmonella Dublin.* A review. J. Hyg. (Lond); 79: 25–38.

Jeffrey M, Hogg RA (1988). Concurrent bovine virus diarrhoea virus and Campylobacter foetus infection in an aborted bovine foetus, Veterinary Record 122: 89–90.

Je-In L, Ill-Hwa K (2007). Pregnancy loss in dairy cows: The contributing factors, the effects on reproductive performance and the economic impact J. Vet. Sci. 8(3): 283–288.

Kim JH, Lee JK, Lee BC, Park BK, Yoo HS, Hwang WS, Shin NR, Kang MS, Jean YH, Yoon HJ, Kang SK and Kim DY (2002). Diagnostic survey of bovine abortion in Korea: with special emphasis on Neospora caninum, J. Vet. Med. Sci. 64: 1123–1127.

Kradel DC (1978). Abortion storms projection for the future. Cornell Vet. 68: 95–199.

Lang GH (1989). Q fever: an emerging public health concern in Canada. Canadian J. Vet. Res. 4(53): 1-6.

Lisak V, Vosta J, Rehacek J (1989). Incidence of Coxiella burnetti and Chlamydia psittaci in cattle in Southern Bohemia. Vet. Med. Praha. 34(7): 403-10.

Lopez-Gatius F, Lopez-Be'jar M, Murugavel K, Pabon M, Ferrer D, Almeri'a S (2004). Neospora-associated abortion episode over a 1-year period in a dairy herd in North-east Spain. J. Vet. Med. B(51): 348–352

Losson B, et Bourdoiseau G (2000). Neospora caninum, un nouvel agent abortif chez les bovins. Bulletin des GTV n°7 107-114.

Lounes N, Bouyoucef A (2009). Les avortements infectieux en Algérie: Enquête sur terrain in: 1eres journées Maghrébines d'épidémiologie Animale, 09-10 mai 2009. www.univ-blida.dz.

McClausland IP, Slee KJ, Hirst FS (1987). Mycotic abortion in cattle. Aust. Vet. J. 64: 129–32.

Mineo TWP, Carrasco AOT, Marciano JA, Werther K, Pinto AA, Machado RZ (2009). Pigeons (Columba livia) are a suitable experimental model for Neospora caninum infection in birds. Vet.

Parasitol. 5: 149-153.

Radostits OM, Leslie KE, Fetrow J (1994). Herd Health: Food Animal Production Medicine (2nd ed.), W.B. Saunders Co., Philadelphia, PA.

Reitt K, Hilbe M, Voegtlin A, Corboz L, Haessig M, Pospischil1 A (2007). Etiology of Bovine Abortion in Switzerland from 1986 to 1995 – A Retrospective Study with Emphasis on Detection of Neospora caninum and Toxoplasma gondii by PCR J. Vet. Med. A(54): 15–22.

Renukaradhya GJ, Rajasekhar M, Raghavan R (1996). Prevalence of infectious bovine rhinotracheitis in South India. Rev. Sci. Tech. 15 (3): 1021- 8.

Thompson G, Canada N, do Carmo Topa M, Silva E, Vaz F, Rocha A (2001). First confirmed case of Neospora caninum -associated abortion outbreak in Portugal. Reprod. Domest. Anim. 36: 309–312.

Thurmond MC, et Hietala SK (1997). Effect of congenitally acquired Neospora caninum infection on risk of abortion and subsequent abortions in dairy cattle, Am. J. Vet. Res. 58: 1381-1385.

To H, Htwe KK, Kako N, Yamaguchi T, Fukushi H, Hirai K (1998). Prevalence of Coxiella burnetti infection in Dairy Cattle with reproductive disorders. J. Vet. Med. Sci. 60(7): 859-861.

W.B. Saunders Co, Philadelphia, PA (1994). (2nd ed.).

Woodbine KA, Medley GF, Moore SJ, Ramirez-Villaescusa AM, Mason S (2009). A four year longitudinal sero-epidemiology of bovine herpes virus type-1 (BHV-1) in adult cattle in 107 unvaccinated herds in South west England. BMC. Vet. Res. Jan. 30; 5(1): 5.