YERSINIA ENTEROCOLITICA AND ITS ROLE IN BOVINE BRUCELLOSIS MONITORING: A STATISTICAL ANALYSIS *

Carla Grattarola [1], Maria Silvia Gennero [1], Simona Zoppi [1], Stefania Bergagna [1], Laura Chiavacci [2] and Alessandro Dondo [1]

***SUMMARY** : 184 animals were tested to detect Yersinia spp. from faeces: 76 of them were positive to RBT and/or CFT and 108 were negative. Association between serological tests and detection of Yersinia spp. was studied considering health status of native herds (PROC. LOGISTIC and PROC. GENMOD – SAS 8.2). Odds ratio value confirms that Yersinia presence increases the chances to detect false seropositivities compared with Yersinia free herds, according to bibliography.*

Keywords: *Y. enterocolitica*, cross reactivity, statistical analysis, brucellosis.

***RESUME** : Cent quatre vingt quatre animaux ont été étudiés pour la recherché de Yersinia spp dans les fèces : 76 ont fourni une réponse positive au test au rose bengale et/ou à la fixation du complément et 108 une réponse négative. L'association entre les résultats des tests sérologiques et la détection de* Yersinia *spp a été étudiée en tenant compte du statut sanitaire des troupeaux (Proc. Logistic et Proc. Genmod-SAS 8.2). Conformément à la bibliographie, la valeur de l'odds ratio confirme que la présence de Yersinia augmente le risque de réponses faussement positives par rapport à des troupeaux indemnes de Yersinia.*

Mots-clés : *Y. enterocolitica*, réaction croisée, analyse statistique, brucellosis.

I - INTRODUCTION

The bovine health situation in the respects for the brucellosis in Piedmont Region has gone progressively improving, becoming certified-free provinces, in the '96 the provinces of Vercelli, Biella and Alessandria, in '97 Asti, Novara and Verbania, and also in the '98 the provinces of Turin and Cuneo. It is important to keep and improve the reached healthy standard to prevent the outbreak of new seropositivity.

The purpose of the present study is to contribute to the diagnosis of the atypical seropositivity observed in Piedmont Region during the year 2004 and to elaborate a diagnostic protocol adequate to the definition of the single reactors.

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[1] Istituto Zooprofilattico Sperimentale di Piemonte, Liguria e Valle d'Aosta, Via Bologna 148, 10154 Torino, Italy; mariasilvia.gennero@izsto.it

[2] Osservatorio Epidemiologico Regione Piemonte

II - MATERIAL AND METHODS

Twenty-one cattle herds characterized by the presence of suspicious serological cross-reaction [Johnson *et al.*, 1994 ; Kittelberger *et al.*, 1995 ; Pouillot *et al.*, 1998] have been identified and tested. On the bovines positive to the Complement Fixation Test (CFT), whereas possible, the complete protocol of diagnostic investigations *in vita* and *post mortem* has been applied, aimed to bacteriological isolation of *Brucella* spp from milk and organs [Alton *et al.*, 1988], of *Yersinia* spp. [Garin-Bastuji, 1999, Godfroid *et al.*, 2002], *Salmonella* spp. and *Escherichia coli 0:157* from faeces and of *Francisella tularensis* from liver [A.A.V.V., 2004]. They have been furthermore considered the results of *Yersinia* spp presence in faeces of regularly slaughtered bovines belonging to certified-free herds (negative control).

III - RESULTS

One hundred and thirty two bovines have been tested, of which 45 seropositive to CFT and 87 cohabitants, of which 58 seronegative and 29 only positive to the Rose Bengal Test (RBT). In all of the cattle herds, epidemiological and diagnostic data have excluded brucellosis infection.

Results of serological examination are shown in table 1.

Other controls, excluding those ones for *Y. enterocolitica*, did not shown any positivity, it can therefore assume any involvement in the seropositivity for *Brucella* in our territory (table 2).

Table 1

Samples positive to CFT

Samples	CFT 20CFU/ml	CFT 40CFU/ml	CFT 80CFU/ml	Total
	29	15	1	45

Table 2

Monitored herds and fecal samples examined to evaluate the presence of cross-reactivity agents *(*monitored herds - **examined fecal samples)*

Y. enterocolitica	***E. coli O:157***	***Salmonella* spp.**	***F. tularensis***
21* (*15 positive herds*)	18* (0)	18*(0)	3* (0)
Y. enterocolitica	***E. coli O:157***	***Salmonella* spp.**	***F. tularensis***
132**(*53 positive samples*)	97**(0)	97**(0)	5** (0)

Regarding to the presence of *Y. enterocolitica* in cattle herds, a statistical analysis has been made to evaluate the association between *Y. enterocolitica* presence and seropositivity to *Brucella*, using the results of the samples coming from seropositive herds and that ones of the samples coming from seronegative herds, using the PROC. LOGISTIC and the PROC. GENMOD of the SAS 8.2 program [Altman, 1991 ; Stokes *et al.*, 2000].

One hundred and eighty four bovines have been tested: out of these 76 were positive to CFT and/or RBT and 108 were negative.

Two statistical models have been used considering relationship between cattle and origin herds:

- Evaluation of association between seropositivity *vs. Brucella* spp. and *Y. enterocolitica* isolation in the same examined cattle;
- Evaluation of association between *Y. enterocolitica* isolation in cattle and seropositivity *vs. Brucella* spp. in the same herd.

IV - DISCUSSION

In case of seropositivity for *Brucella*, having excluded previously the infection by direct methods (bacteriological test and molecular biology techniques), false positive to serological test can be explained by the presence of *Y. enterocolitica* in faeces.

Therefore, *Odds Ratio* value confirms that *Yersinia* presence increases the chance to detect false positive samples compared with *Yersinia*-free herds, according to bibliography [Johnson *et al.*, 1994 ; Kittelberger *et al.*, 1995 ; Pouillot *et al.*, 1998].

However, it could have caused himself a distortion of the assessment, because the sampling has not been homogeneous (samples coming from seronegative herds generally turned out 1:1 respect to the membership stock farm, while the samples, belonging to seropositive stock farms have turned out 1:5).

Searching other interfering agents, as described in literature, does not seem to be useful to diagnose aspecific seropositivity *vs. Brucella* spp.

Therefore we consider as opportune, in the future, for a quick definition of dubious cases to limit the controls, *in vita*, to the *Brucella* spp isolation in milk, if possible, and to *Y. enterocolitica* in fecal samples of seropositive cattle and at least 5 cohabiting animals.

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