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Excretion of *Coxiella burnetii* by dairy cows: consequences for disease screening and control

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Reliable detection of *Coxiella burnetii* shedders is a critical point for the control of the spread of the bacterium between animals and from animals to humans. *Coxiella burnetii* is shed by ruminants mainly by birth products, but may be shed also by vaginal mucus, milk, and faeces, urine and semen. However, the informative value of these types of samples to identify shedders under field conditions is unknown. Our aim was then to describe the responses obtained using a real-time PCR technique applied to milk, vaginal mucus and faeces samples taken from 280 dairy cows in five commercial dairy herds known to be naturally infected with *Coxiella burnetii*, and to assess their putative associations. Cows with only one type of sample found positive accounted for more than 85% of apparent shedders. By contrast, cows with positive results for all three samples were scarce (less than 2%). Testing a cow based on only one type of biological sample may lead to misclassify her towards *Coxiella burnetii* shedding and then to underestimate the risk of bacterial spread within a herd and of transmission from animals to humans.

Warning network on bovine Salmonellosis in Burgundy: results after 10 years

E. Petit

In 1996, whereas a wave of clinical episodes of bovine salmonellosis prevailed in Burgundy, the GDS of Burgundy proposed at the departmental laboratories of analyses and at the veterinary surgeons to set up a regional warning network on bovine salmonellosis. For any Salmonella isolated on bovines by the departmental laboratories of Burgundy, the system transmits by the way of the GDS Regional Federation to the veterinary practices of the concerned region information available on the identified case. In 10 years, the network has send nearly 1 000 warnings addressed to 10 000 veterinary practices. The multiannual assessment reveals a constant decrease of the number of cases since 1996, which is also observed in other networks like the Epidemiosurveillance system of bovine salmonellosis (RESSAB). The geographical distribution of the cases appears very variable one year on the other and shows sometimes micro local outbreaks. Some others informations, like the serovar, the antibiotic resistance profile and the observed clinical signs, are also collected. Thus, *Salmonella typhimurium* prevails regularly with more than 80% of isolated strains. This system presents some defaults on representativeness of cases, but its main goal is the vigilance of vets, and it constitutes an active and cheap regional epidemiosurveillance system.

Evaluation by simulation of the strategy of culling PI animals in cattle dairy herds infected by the BVDV

Pauline Ezanno, A. Joly, Christine Fourichon & H. Seegers

Control strategies have been implemented to limit the spread of infections with the bovine viral diarrhoea virus (BVDV), such as test-and-cull of persistently infected (PI) animals. Our objective was to study the influence of the survival of PI animals on the spread of BVDV in cattle dairy herds. A compartmental stochastic discrete time model has been developed, that included herd dynamic and BVDV infection dynamic. In the model, the herd was divided into 5 groups: calves, young heifers, older heifers, lactating cows and dry cows, representing a typical French dairy cattle herd. Several infection states were considered: protected by maternal antibodies (M), susceptible (S), transiently infected (T), immune (according to the infection time: not pregnant (R), in early (Ra), in mid- (Rb), or in late pregnancy (Rc)), PI (P). PI animals had a higher mortality rate that other animals. The model sensitivity to variation in the survival of PI animals was analysed, the virus being introduced once in the herd by the introduction of infected animals. Without any intervention, the half-life of PI animals is estimated to be one year. Shorter half-life (1, 2, 3 and 6 months) was tested, simulating an early exit resulting from the test-and-cull. The studied model outputs were the probability of infection extinction 6 months, 1 or 2 years after the virus introduction, the time needed to reach a probability of infection extinction of 50, 70, 80, 90 or 100%, and the epidemic size (mean cumulative numbers of PI and transiently infected animals) 10 years after virus introduction. The results were very sensitive to a decrease in survival of PI animals. One year after virus introduction, the infection extinction occurred on average for 40% of the herds if none intervention was done. For a half-life of PI animals decreased to 6, 3, or 1 month, extinction occurred for 60, 70 and 90% of the herds, respectively. The infection extinction occurred for 90% of the herds after more than 5 years without intervention. On average, it occurred before one year for a half-life of PI animals decreased down to 1 month. The epidemic size decreased when reducing the half-life of PI animals, with 35 to 65% less PI animals and 30 to 80% less transiently infected animals. Our study allowed to quantify the virus clearance speed in cattle dairy herds structured in groups according to the ability to early detect and cull PI animals.

BSE study by discriminant analysis

E. Morignat, Anne-Gaëlle Biacabe, C. Ducrot, T. Baron & D. Calavas

A retrospective biochemical study of the BSE cases diagnosed in France allowed to identify, besides the classical form, two new atypical forms of BSE. These different forms are identifiable thanks to different electrophoretic profiles of the prion protein in Western Blot. This paper presents the use of the Fisher Discriminant analysis to differentiate the three molecular types of BSE. This quantitative method takes into account all the biochemical parameters in order to classify the different forms of BSE cases.

Blue tongue surveillance in France and in the west Mediterranean area

G. Gerbier, J. Parodi, Fabienne Biteau-Coroller, T. Baldet, B. Mathieu, S. Zientara, Catherine Cetre-sossah & F. Roger

A summary of the knowledge acquired on bluetongue since 2000 is presented. After the successive introductions of three different serotypes in Corsica, it is possible to compare the impact of each strain and to analyse the ways of propagation. The various strategies of surveillance (clinical, serologic and entomological) in place between 2000 and 2006 in Corsica and on the continent are analyse according to the risk of introduction or the statute of the area (infected or free). The effectiveness of the generalized vaccination campaigns and the situation in the neighbouring countries are also discussed.

Feed infection for BAB cases of BSE. Complementary studies on bovine feedstuffs period of consumption and use of poultry feedstuffs in farms

Nathalie Jarrige, C. Ducrot, Géraldine Cazeau, E. Morignat & D. Calavas

A case/control study [Jarrige *et al.*, submitted] on Bovine Spongiform Encephalopathy (BSE), showed that feeding cattle with proprietary concentrates before the age of two was the main risk factor to explain infection of born after the ban of meat-and-bone meal, in force since 1990 in France. At a lesser extent, consumption of milk replacers by bovines, and poultry proprietary concentrates purchase was also identified as a risk factor. Using the same data, two complementary studies were carried out, one taking into account the possible infection periods for cattle, the other restricting the sample to the farms where poultry was present. A conditional logistic regression model was used, adjusting for herd size and cattle production type. That study showed that cattle consumption of proprietary concentrates before the age of two or before and after the age of two were significant risk factors (OR=7.8 [1.4-45.3] and OR=6.8 [1.2-37.9], respectively). On the other hand, consumption of bovine proprietary concentrates only after the age of two did not appear significant. In addition, in farms having poultry, feedstuff purchase for these species was also significantly associated with BSE occurrence (OR=3.9 [1.1-13.7]).

Evaluation of the seroprevalence of caprine herpesvirus 1 infection in South-West of Europe

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Serums from more than 1200 goats distributed through Belgium, Spain, France and Italy have been analysed by sero-neutralisation in order to update the epidemiological situation of the infection of goats by caprine herpesvirus 1 (CpHV-1). The serological analyses have shown that CpHV-1 is not likely to be present in Belgium and France whereas a high apparent prevalence is observed in Spain and Italy.

Trend of *Brucella suis* infection in wild boar in Piedmont Region (2002-2005)

Maria Silvia Gennero, Carla Grattarola, Stefania Bergagna, Simone Zoppi, A. Barbaro & A. Dondo

Brucellosis caused by *Brucella suis* is a zoonosis common to different animal species, including the wild boar. Due to the numerous wild boar population in Piedmont, since 2000 a monitoring programme was implemented to check their health status, wherein serological monitoring for Brucellosis is one of the programme targets. After serological positivity to the infection was found in some herds in 2001, typing of the infectious agent was performed by bacteriological examination (BE). Serum samples from 2688 wild boars were examined by the Rose Bengal Test (RBT) and 3535 by the Complement Fixation Test (CFT); samples from 1864 animals were analysed by bacteriology. In 160 animals *Brucella spp.* was isolated by bacteriological determination. *Brucella* infection may be considered endemic in some areas in Piedmont Region.

Description of sheep diseases in the district of Alaotra lake (Madagascar) using participative epidemiology

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Between 1997 and 1998 an epizootic of African Swine Fever did cut by half the number of pigs within the region of Alaotra lake (Madagascar) which was initially of 40 000 animals. To face this outbreak, the farmers of this region decided to turn to ovine farming, activity which had remained marginal. Because the sanitary conditions of this farming sector were unknown or uncertain at this stage, the use of participatory epidemiological tools appeared most adapted to include the population in the establishment of diagnosis, the formulation of solutions and their implementation in adequacy with the socio-economic context. According to the farmers, the sanitary situation was greatly determined by the internal parasitic diseases (78%), their distribution differentiated according to of the type of environment. They were responsible for 88% of the mortalities whereas the infectious diseases (viral or bacterial) or the external parasitic diseases were responsible respectively for only 10 and 2% of the mortalities. From this participatory epidemiological study, three ways of improving the sanitary situation were identified and initiated: the establishment of a medical prophylaxis calendar and the amelioration of both the habitat and the alimentation.

Estimation of the global cost of an IBR control program in Belgium

M. Dispas, J-Y. Houtain, B. Muylkens, B. Gauthier, P. Kerkhofs & E. Thiry

The estimation of the global cost of a control program of IBR in Belgium is realized for a period of 10 years. Based on the protocols for the acquisition and maintaining of the sanitary status, that will be defined in the forthcoming legislation, and a constant decrease of the IBR prevalence, the annual cost by status, the mean individual costs by certification step and the mean individual costs according to the herd size are calculated. The less costly status to acquire and maintain is the IBR-free status.

Is the Walloon European Wild boar a potential reservoir of Pseudorabies virus for porcine livestock?

G. Czaplicki, J. Dufey & C. Saegerman

With the screening organized for the monitoring of the classical swine fever in wild boars in Walloon area, a serologic investigation concerning the SuHV1 virus was undertaken between September 2004 and December 2005 into approximately 2 300 wild boars. This survey is based on in-house commercial Elisa test validated for wild boar. This study shows the existence of an endemic infection of the Walloon wild boars by Pseudorabies virus, with important sub-regional disparities. The preliminary analysis of various parameters (age, sex, season or month of sampling), according to the biology of wild boars, makes it possible to wake up or confirm tracks concerning the pathogenesis of the infection in this species.

Ten years of molecular epidemiology of bovine tuberculosis in Belgium

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After decades of control and eradication programs, the herd incidence of bovine tuberculosis diminished progressively under the 0,1% threshold since 1997. Currently, bovine tuberculosis due to *Mycobacterium bovis* is still sporadic. Since Belgium was recognised officially free of bovine tuberculosis in 2003 (decision 2003/467/CE), 5 to 10 infected herds are identified yearly. In order to better understand the epidemiology of the infection in our country, the molecular typing of the strains of *M. bovis*, isolated since 1995, was done. Between August 1995 and November 2005, the isolation of *M. bovis* was done in 233 cattle herds. The isolates of *M. bovis* originating of 93% of these infected herds were typed by restriction techniques fragment length polymorphism (RFLP IS6110) and spoligotyping. Each of these techniques presents a different discriminatory power according to the genotypes, but the two techniques are complementary. Together, they distinguish more than 40 genotypes. The technique of mycobacterial interspersed repetitive unites - variable number tandem repeat analysis (MIRU-VNTR), evaluated on a sample of 128 isolates, differentiate the *M. bovis* strains, similarly to the two other techniques implemented together. On the other hand, the stability of the profiles obtained by the MIRU-VNTR technique was shown on a sample of isolated strains originating from the same cattle flock and in herds presenting documented epidemiological links. This technique, easy to implement and highly performant, will replace progressively the two others typing techniques. Between 1995 and 2005, 12 lineages with distinct genotypes were observed in Belgium. A lineage is clearly dominating since it represents 48% of the infected herds. This lineage was associated with the new peak of incidence of bovine tuberculosis observed in the province of Liège between 1995 and 1996. Other lineages are more rarely observed (maximum 9% of the infected herds). A retrospective analysis shows that certain lineages can reappear several years after having been observed and that new type of strains appears punctually. Two type of strains that were never observed since 1995 appeared in 2004. This result suggests that in addition to the circulation of strains of *M. bovis* between Belgian flocks, other ways of introduction of bovine tuberculosis must be suspected in certain case. The continuous molecular typing of the isolated strains of *M. bovis* constitutes a precious tool for the re-orientation of the epidemiological investigations leading to take appropriate management measures.

Estimating disease prevalence and diagnostic test characteristics using a Bayesian approach

N. Praet, P. Dorny, C. Saegerman, T. Marcotty & D. Berkvens

Estimating the prevalence of a disease from the results of one or more diagnostic tests applied to individuals of unknown disease status invariably means that, in the absence of a gold standard and without external constraints, more parameters must be estimated than the data permit. The Bayesian approach allows the use of expert knowledge to reduce the number of parameters to be estimated to the number of estimable parameters and permits estimation of the true prevalence of the disease and of characteristics of the tests involved in the diagnosis. A Bayesian statistical model assuming conditional dependence has been developed and applied to results of four tests for the diagnosis of porcine cysticercosis in a population of 868 Zambian village pigs. Several validation criteria were checked. The true prevalence of the disease in this population was estimated to be around 60%, three times higher than previously reported in this area. The Bayesian approach seems to be a reliable technique for modelling multiple test results for the diagnosis of a disease. Its application can be extended to the diagnosis of other diseases using a variable number of tests.

Milk production and BSE risk in France. Analysis at the farms level

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Following the epidemic of Bovine Spongiform Encephalopathy (BSE) in the UK which culminated in 1993, France was among the first BSE affected countries in continental Europe. As in the UK, dairy cattle were much more affected than beef suckler cattle, because of their higher consumption of concentrates. However, the effect of the average milk yield at the herd level has not been examined in relationship with the risk of BSE. The paper deals with that question. Two databases were merged: the French Milk Recording Database with over 70 000 herds recorded every year, and the French BSE database. Cases detected since July 1, 2001 were considered, when systematic tests were implemented, in order to avoid any notification bias. Only Holstein herds were considered, because their distribution across the French territory is fairly homogenous as compared to the other breeds. Regarding the BSE risk, odds ratios for each class of milk production were estimated, by the use of logistic regression models with proper adjustments on factors such as the farm location, herd size and year of milk record. The analysis was carried out on 33 584 herds with milk yield data available during the period of interest, *i.e.* the year of birth of the cases: among them, 144 were affected with BSE between July 2001 and July 2003. The main result was a 3.8 folded enhanced BSE risk for the top class of highly producing herds (standard milk yield of 10 000 kg and above), by reference to the least producing herds (< 7000 kg). The BSE risk was found to be uniform for the vast majority of herds for which the average milk yield was between 7000 and 10 000 kg. The herd size was not linked to the BSE risk apart from the enhanced risk of discovering at least one affected animal when the number of animals increases.