SOCIETY FOR VETERINARY EPIDEMIOLOGY AND PREVENTIVE MEDICINE

Proceedings of a meeting held at

Martigny, Switzerland

on the 24th, 25th and 26th March 2004

Edited by S.W.J. Reid, F.D. Menzies and A.M. Russell

ISBN 0 948073 64 0

ACKNOWLEDGEMENTS

Secretarial assistance in collating the proceedings was provided by Linda Braida and Martina Monaghan.

The following bodies provided financial support for the conference and the publication of these proceedings:

Companies represented by the Swiss Pharmaceutical Contact Group for Research Questions (KGF) that contributed financially:

Novartis **Roche** **Serono** **Syngenta**

Cantonal Veterinary Office (Bern)

Prionics AG

Swiss Federal Veterinary Office

Swiss Forum for Epidemiology & Animal Health

Veterinaria AG

CONTENTS

TOOLS

COMPANION ANIMALS

POLICY

ENDEMIC DISEASES

ANTIMICROBIALS

ANIMAL HEALTH ECONOMICS

ANIMAL MOVEMENT

***

Society for Veterinary Epidemiology and Preventive Medicine

Past Presidents
Executive Committee Members & Life Members
Gareth Davies Lectures & Opening Conference Plenary Lectures
Membership Application Form & Treasurer’s Address
Constitution and Rules

TOOLS

SPATIO–TEMPORAL ANALYSIS OF THE 1999-2000 AVIAN INFLUENZA EPIDEMIC IN THE MAIN POULTRY PRODUCTION AREA IN NORTHERN ITALY

A. MANNELLI[1], N. FERRE, S. MARANGON AND M. DRIGO

SUMMARY

Spatio-temporal analysis, logistic regression, and survival analysis were used to study the highly pathogenic avian influenza (HPAI) epidemic of 1999-2000 in northern Italy, in one of the most important poultry production areas in Europe. After circulation of low pathogenic virus, HPAI was first detected in early December of 1999. The epidemic peaked between late December and the first decade of February 2000. Multiple areas of early outbreak insurgence were detected in the provinces of Verona (Veneto) and Mantova (Lombardy). The provinces of Bergamo and Milano (Lombardy) were affected by a later, north western spreading of the infection. Space-time K function suggested clustering of outbreaks occurring within ~ 20 day from one another, and logistic regression analysis showed that inter outbreak distances < 3 km were associated with relatively short inter outbreak times. Taking into account the time of most likely outbreak insurgence after viral transmission from an infected premise (IP), we found that proximity (within 4 km) from an IP was associated with HPAI risk (population attributable fraction = 52.9%) and was the most important predictor in a Cox regression model. Moreover, hazard was greater for turkeys than for other bird species, and for flocks in Lombardy region as compared to Veneto.

INTRODUCTION

Highly pathogenic avian influenza (HPAI) is caused by H5 and H7 subtype viruses, and causes heavy losses in poultry production, due to morbidity and mortality approaching 100%, and to strict disease control measures (Alexander, 2000). HPAI viruses originate from low pathogenic avian influenza strains (LPAI), and mutation is favoured by intense viral circulation in domestic bird populations, after transmission from wild reservoirs. Structure and organization of poultry industry are mainly dictated by economic convenience, whereas biosecurity and communicable disease prevention are given scarce consideration (Capua & Marangon, 2000). In Italy, ~ 65% of poultry production is concentrated in the northern regions of Lombardy and Veneto, and up to 70000 birds/km^2 are reared in some municipalities of the province of Verona.

In March 1999, LPAI virus (H7N1 subtype) was reported from northern Italy. Severe measures were however not adopted to control this infection because only HPAI is considered in the official EU definition of avian influenza. In December 1999, HPAI was detected in a turkey farm in the province of Verona and, in the subsequent four months, a devastating epidemic

---

[1] Alessandro Mannelli, Facoltà di Medicina Veterinaria, Università di Torino, via Leonardo da Vinci, 44, 10095 Grugliasco (To), Italy. Email: Alessandro.Mannelli@unito.it

caused the destruction of ~ 16 million birds belonging to all intensively reared species, such as turkeys, chickens, guinea-fowls, and game birds. Following the 1999 – 2000 HPAI epidemic, major changes in the organization and structure of poultry industry in northern Italy have been proposed, as well as contingency plans in the case of future epidemics (Capua et al., 2002). Under these circumstances, epidemiology provides scientific evidence to support measures in preventive veterinary medicine. Spatial, and spatio – temporal analysis was previously integrated with classical epidemiological techniques to study infectious disease epidemics in livestock (Wilesmith et al., 2003). In this study, we used such tools to explore patterns of the 1999 – 2000 HPAI epidemic. A spatially meaningful predictor was then used in survival analysis to quantify the adjusted effects of proximity to previous outbreaks, bird species, and region on HPAI hazard for poultry flocks.

MATERIALS AND METHODS

Spatio – temporal analysis

Spatial distributions of poultry flocks, and of HPAI outbreaks in 1999 – 2000 in Lombardy and Veneto were visually explored through contour maps of two – dimensional kernel density estimates, using the *kde2d* function, package MASS, in the R software ver. 1.8.0 (Ihaka & Gentleman, 1996, Venables & Ripley, 1999). Spatio – temporal pattern was visualised by linear interpolation of date of outbreaks using the akima package, and by space – time K function ($K_{s,t}$) using SPLANCS (Rowlingson and Diggle, 1993). $K_{s,t}$ is the cumulative count of other outbreaks within distance *s* and time *t* from an outbreak. The difference (D) between $K_{s,t}$, and the product of space (K_s) and time (K_t) K functions is a measure of space – time interaction in outbreak occurrence. Peaks in the D surface diagram were interpreted as departures from random space – time distribution of outbreaks. Space – time interaction was then tested by logistic regression, using outbreak pairs as units of analysis. Inter outbreak distance < 3 km was the model outcome, whereas quartiles of inter event time were included as predictors (SAS, 1999).

In order to quantify the importance of spatial proximity to infected premises (IP) in the risk of AI at the population level, we classified farms based upon frequencies of IP's in temporal risk window (TRW) that were located within increasing distances from each farm (Taylor et al., 2003). TRW for an IP is the time interval during which clinical signs of disease can be detected in a flock due to transmission from the IP. In our calculation, we used two and six days as minimum and maximum incubation period for AI. Therefore, TRW began one day after the detection of first clinical signs in an IP, and ended six days after IP's destruction. The population attributable risk associated with the presence of IP's within a certain distance from a farm was obtained by SAS® ver. 8 and R.

Survival analysis

Multivariate survival analysis by Cox regression was used to evaluate the effects of proximity to IP's in TRW, bird species, and region on the hazard of HPAI for poultry flocks. Times of observation were both right and left censored. Right censoring corresponded to regular slaughtering of birds or end of the study period, whereas left censoring was due to stocking of flocks after the beginning of the study period. The ENTRY option in PROC PHREG (SAS, 1999) was used to allow exclusion of left censored observations from the risk set before the time of flock stocking. Interaction terms between predictors were retained in the final model if significant at the 10% level. The BASELINE option in PHREG was used to obtain survivor function estimates. The data set COVARIATE was created containing the values of explanatory variables for which survivor functions were estimated.

RESULTS

Spatial distributions of poultry farms and HPAI outbreaks from 5 December 1999, to 5 April 2000 are shown in Figures 1, 2. The epidemic occurred in the western part of the area of highest flock concentration, and involved 230 out of 1159 (19.8%) flocks in Lombardy, and 152/2102 (7.2%) in Veneto. Meat turkey flocks were heavily affected (175/798, 21.9%). Most of the outbreaks occurred between late December 1999 and the first decade of February 2000 (Figure 3). Areas of early epidemic insurgence were detected in the provinces of Verona (Veneto) and Mantova (Lombardy), as shown in Figure 4, whereas outbreaks in the provinces of Milano and Bergamo (Lombardy) were the results of later, north western spread of the infection.

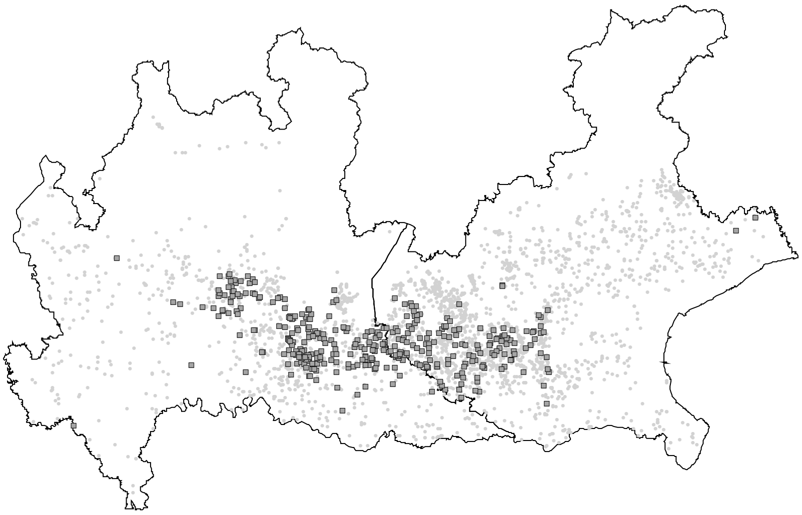


Fig. 1 Distribution of poultry farms (grey squares) in the northern Italian regions of Veneto (right) and Lombardy (left). Black squares indicate locations of highly pathogenic avian influenza (HPAI) outbreaks that occurred during winter 1999 – 2000.

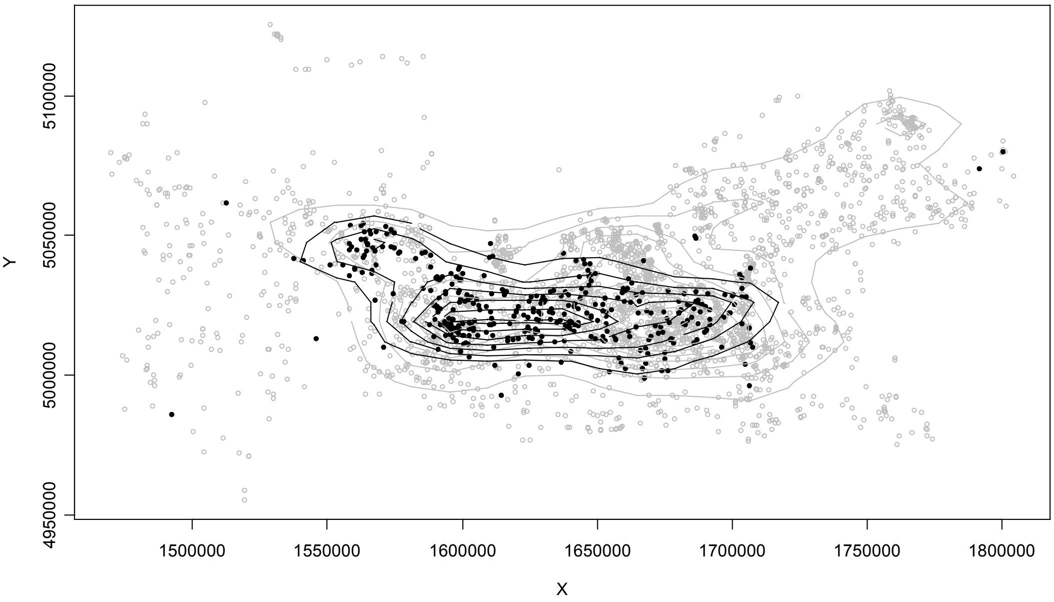


Fig. 2 Contour map of bivariate kernel density estimates, and point locations of poultry flocks (grey) and HPAI outbreaks (black) in Veneto and Lombardy, in 1999 – 2000. X and Y coordinates are in the Gauss Boaga system.

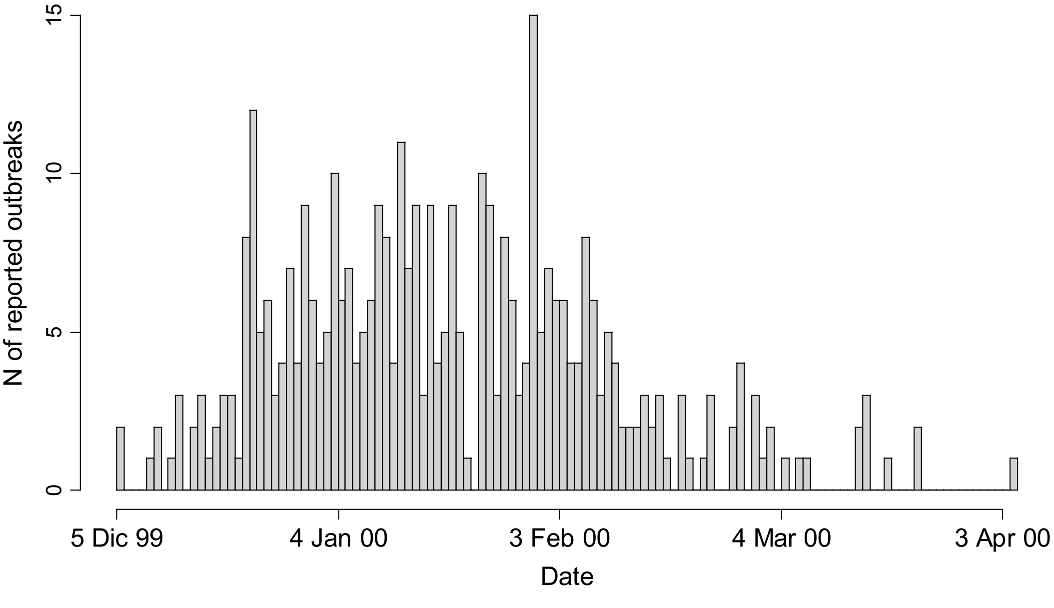


Fig. 3 Distribution of the number of HPAI outbreaks by day of detection of clinical signs, in northern Italy in winter 1999 – 2000.

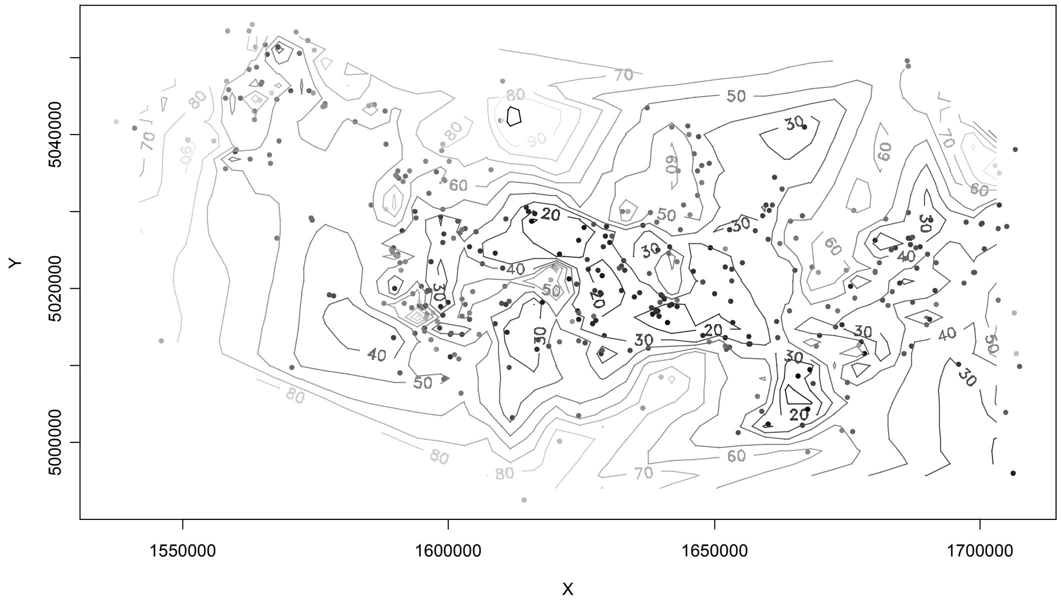


Fig. 4 Contour map of linear interpolation of date of HPAI outbreak occurrence and point locations, in Veneto and Lombardy. Line label numbers indicate days from 5 December 1999. Grey intensity indicates date of outbreak: black colour corresponds to locations of early occurrence of outbreaks, whereas late spreading of the epidemic is indicated by light grey lines and points.

Spatial – temporal K function showed a major pattern of clustering for outbreaks occurring within ~ 20 days from one another (Figure 5). The logistic regression model for space – time association was significant (likelihood ratio $\chi^2 = 172.5$, 3 df, $P<0.001$) and showed no evidence of poor fit (Hosmer and Lemeshow test: $\chi^2 = 0.0$, 2 df, $P = 1.0$). Short inter event times were associated with short inter event distances. In fact, outbreaks occurring within 10 days from one another were more likely to be separated by distances < 3 km, as compared to outbreaks separated by longer times (Tab. 1).

Population attributable fraction for the presence of at least one IP in TRW within 4 km from a flock was 52.9%, we therefore included a corresponding dichotomous variable among predictors in survival analysis. The final Cox regression model included proximity to IP, species (turkey vs other bird species), region (Lombardy vs Veneto), and turkey by proximity interaction (Table 2). Likelihood ratio test was significant ($\chi^2 = 375.5$, 4 df, $P<0.001$). The hazard of HPAI was greatest for flocks situated within 4 km from IP's. Moreover, hazard was greater for turkeys than for other bird species, and for flocks in Lombardy region as compared to Veneto. A significant, negative interaction indicated that the effect of proximity to IP was stronger for other species than for turkeys. Survivor estimates are shown in Figure 6.

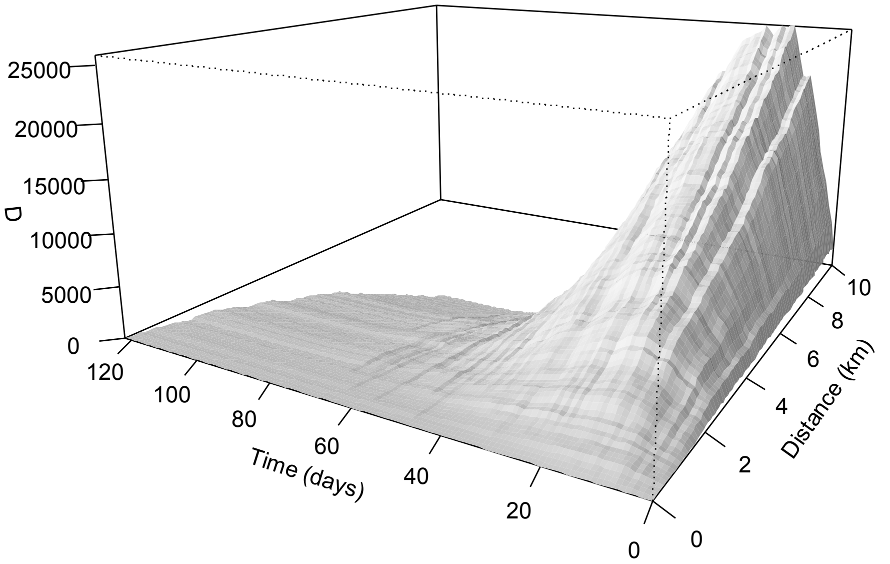


Fig. 5 Spatio – temporal K function of HPAI outbreaks in Veneto and Lombardy, in winter 1999 – 2000. Peaks in D surface indicate interaction between space and time of outbreak occurrence.

Table 1. Logistic regression analysis of space – time association of the occurrence of outbreaks of avian influenza in poultry farms in Lombardy and Veneto regions, Italy, in winter 1999 – 2000. Outbreak pairs were units of analysis, and inter outbreak distance <3 km was the outcome.

INTER EVENT TIME	ODDS RATIO	95% CI
<10 d vs >35d	3.9	3.1-5.0
10 – 19 d vs > 35 d	2.2	1.7-2.8
20 – 35 d vs > 35 d	1.4	1.1-1.9

Table 2. Results of the Cox proportional – hazard regression of the HPAI hazard for poultry farms in Veneto and Lombardy, from 21 December 1999 to 11 February 2000.

FACTOR	HAZARD RATIO	95% CI
IP4km*	8.3	5.9-11.7
Turkey vs other bird species	6.9	4.7-10.0
Lombardy vs Veneto region	2.2	1.7-2.7
IP4km x Turkey interaction	0.33	0.21-0.53

* being within 4 km from at least one infected premise in temporal risk window

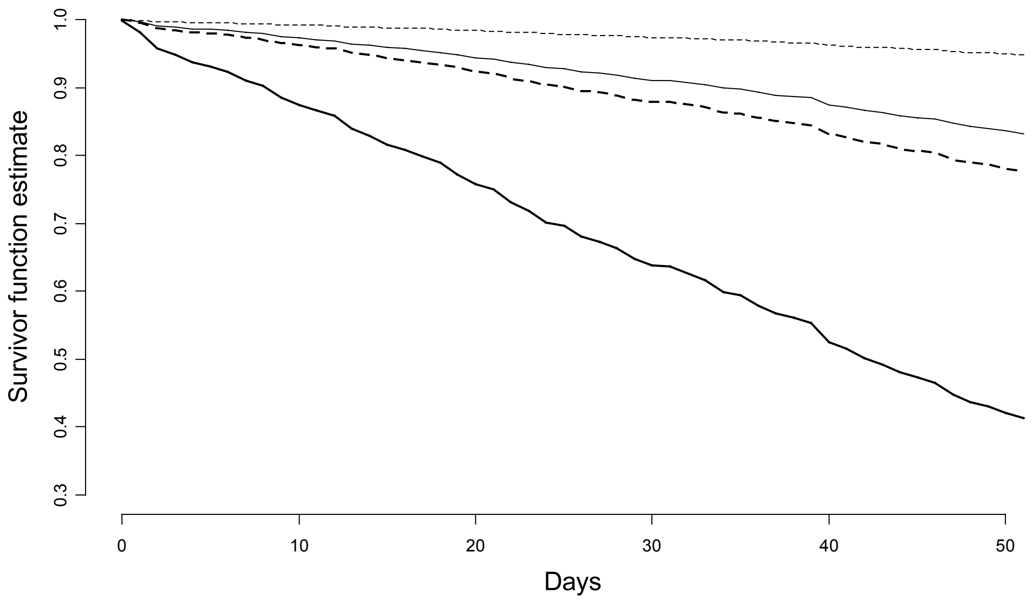


Fig. 6 Survivor function estimates of poultry flocks during the peak of HPAI epidemic, from 21 December 1999 to 11 February 2000. Solid lines indicate turkey flocks, and dashed lines other bird species. For both species categories, bold lines are relative to flocks that were located within 4 km from infected premises in temporal risk window, whereas thin lines represent flocks at longer distances from infected premises.

DISCUSSION

In this study, we used exploratory tools for spatio – temporal patterns in the 1999 – 2000 HPAI epidemic, and included a spatial component in survival analysis, taking into account the most likely time of outbreak detection following viral transmission (TRW) from an IP to another flock. The resulting flock classification variable was the most important predictor of HPAI hazard, together with bird species and region. Flocks within 4 km from a previous outbreak (IP) were at greatest risk. Viral transmission over longer distance also occurred. Moreover, the effect of proximity was stronger for other bird species than for turkey, as shown by a negative interaction term. Turkey flocks, unlike those belonging to other species, were characterised by a relatively high risk of HPAI even when apart from IPs, confirming the high susceptibility of this species. In devising preventive measures based on results of this study, the complexity of the entire system of poultry industry, as well as the ecological dynamics of avian influenza should be considered. A major effort will be required to combine economic priorities with the long term needs of biosecurity and disease control. The zoonotic potential of HPAI makes the above issue even more urgent.

ACKNOWLEDGEMENTS

This study was supported by the Italian Ministry of Health, Department of Food and Nutrition, and Veterinary Public Health. The authors thank Giorgio Zanardi, Istituto

Zooprofilattico Sperimentale della Lombardia e dell’Emilia Romagna, and Emanuela Rossi, Istituto Zooprofilattico Sperimentale delle Venezie, for advice and technical assistance.

REFERENCES

Alexander, D.J. (2000). A review of avian influenza in different bird species. Vet. Microbiol. 74, 3-13

Capua, I. and Marangon, S. (2000). Avian influenza in Italy (1999-2000): a review. Avian Pathol. 29, 289-294

Capua, I., Mutinelli, F., Dalla Pozza, M., Donatelli, I., Puzelli, S. and Cancellotti, F.M. (2002). The 1999-2000 avian influenza (H7N1) epidemic in Italy: veterinary and human health implications. Acta Tropica 83, 7-11

Ihaka, R., and Gentleman, R. (1996). R: a language for data analysis and graphics. J. Computat. Graph. Stat. 5, 299-314

Rowlingson, B.S. and Diggle, P.J. (1993). SPLANCS: spatial point pattern analysis code in S-PLUS. Comput. Geosci. 19, 627-655

SAS Institute Inc. (1999). SAS/STAT® User’s Guide, Version 8, Cary, NC

Taylor, N.M., Honhold, N., Paterson, A.D. and Mansley, L.M. (2003). Quantification of the risk of foot and mouth disease associated with proximity in space and time to other infected premises. Proceedings of Society for Veterinary Epidemiology and Preventive Medicine, Warwick, UK, 195-207.

Venables, W.N., and Ripley, B.D. (1999). Modern applied statistics with S-Plus, Second Edition. Springer – Verlag, New York

Wilesmith, J.W., Stevenson, M.A., King, C.B. and Morris, R.S. (2003). Spatio – temporal epidemiology of foot-and-mouth disease in two counties of Great Britain in 2001. Prev. Vet. Med. 61, 157-170

A BOOTSTRAPPING METHOD FOR DIAGNOSTIC TEST EVALUATION AND PREVALENCE ESTIMATION

J. BEREZOWSKI[1], D. RENTER AND R. EVANS

SUMMARY

A bootstrapping method for tests producing continuous results was developed for estimating the apparent prevalence in samples with varying true prevalence. The method was used to evaluate the accuracy of different cut-off values and to assess Rogan and Gladen's (1978) method for true prevalence estimation. Ordinary least squares regression equations were estimated for the relationship between apparent and true prevalence and used for true prevalence estimation. Bootstrapping/regression produced estimates of true prevalence that were accurate for all cut-off values. An example using an ELISA test for malignant catarrhal fever virus antibodies is given.

INTRODUCTION

The objective of many disease surveys is to estimate the true prevalence of infections or exposures to infectious agents within defined populations. Disease surveys are often based on testing subjects sampled from the population and, in many cases, the screening test detects antibodies or other surrogates for the infectious agent. The validity of prevalence estimates depends on the sampling method and the accuracy of the screening test (Greiner & Gardener 2000b).

The accuracy of screening tests is usually estimated in separate validity studies in which a test is applied to known infected and uninfected subjects. Since screening tests often produce continuously distributed test values, cut-off values must be selected that can accurately classify infected subjects as test positive and uninfected subjects as test negative. The sensitivity and specificity of the test will depend directly on the cut-off value selected and will vary as the cut-off value is increased or decreased. Methods such as receiver operating characteristic (ROC) and two-graph receiver operating characteristic (TG-ROC) analyses are often used to select an optimal cut-off value for the test (Greiner & Gardener, 2000a).

Since screening tests are rarely completely accurate, the test positive or apparent prevalence from survey samples is often adjusted to estimate the true prevalence within the sample. Rogan and Gladen's (1978) method uses the sensitivity and specificity estimates from a validity study and the apparent prevalence from the sample to estimate the true prevalence within the sample. Adjusting the apparent prevalence with sensitivity and specificity estimates implies two assumptions. The first is that the true status of the known infected and uninfected subjects in the

---

[1] John Berezowski, Agri-Food Systems Branch, Alberta Agriculture, Food and Rural Development, 116B OS Longman Building, 6909-116 St, Edmonton AB. T6H 4P2, Canada

validity study were known. The second is that the distributions of test values in the samples of known infected and uninfected subjects of the validity study were similar to the distributions of test values found in infected and uninfected subjects within the sample being surveyed.

Rogan and Gladden's method is not defined for tests where the sensitivity plus specificity equals one and it is censored for prevalence values less than 0.0 and greater than 1.0. Rogan and Gladden prevalence confidence interval estimation is based on a normal approximation with the variance estimated using the number of subjects in the survey sample and the validity study combined. However, the normal approximation is inappropriate for small samples and extreme prevalence proportions (Greiner & Gardener, 2000a).

Bootstrapping is a valid method for estimating the true value of parameters in populations from their values in samples (Efron & Tibshirani, 1993). The validity of bootstrapped estimates is not dependent on sample values conforming to a specified distribution. However, parameter estimate validity is dependent on the samples being random or at least representative samples of population (Efron & Tibshirani, 1993). If the true infection status of validity study subjects is known, and these subjects are representative of infected and uninfected individuals in the population, then it is possible to use the test values from the validity study and bootstrapping to make inferences about test accuracy and unbiased estimates of population true prevalence.

The purpose of this paper is to present a new bootstrapping method for evaluating the accuracy associated with various cut-off values and estimating the true prevalence in survey samples using test values from validity studies and the apparent prevalence from surveys.

MATERIALS AND METHODS

Diagnostic test

The diagnostic test used was a competitive inhibition enzyme linked immunosorbent assay (c-ELISA) that detects antibodies produced against an antigenic epitope that is conserved among a group of closely related gamma herpesviruses called the malignant catarrhal fever (MCF) group of viruses (Li et al., 2001). Optical densities (OD) for each sample were converted to percent inhibitions (PI), which were the ratio of the OD of each sample to the mean OD of the negative controls present on each ELISA plate. In order to avoid negative values a value of one was added to each PI.

Samples

We obtained 132 serum samples from American bison (*Bison bison*) that were uninfected with MCF virus and 128 samples from bison that were infected. Sera were tested with the c-ELISA using previously reported methods (Li et al., 2001).

Assumptions

Our bootstrapping model assumed that the test values came from subjects that were truly infected or uninfected. A further assumption is that these subjects and their test values were representative of infected and uninfected subjects that would be found in populations in which we wish to estimate the true prevalence. In other words, the distributions of test values from the samples of known infected and uninfected subjects would be similar to the distributions of test values found in infected and uninfected subjects in populations with unknown prevalence.

Bootstrapping to estimate apparent prevalence and evaluate the c-ELISA cut-off value

The bootstrapping method was based on producing hypothetical bootstrapped samples by randomly selecting test values (with replacement) from samples of known infected and uninfected test values. If the assumptions of the model remain true, each bootstrapped sample can be considered representative of a single sample, randomly drawn from a population for which the true prevalence was defined by the proportion of test values that were randomly drawn from the sample of known infected subjects. Within each bootstrapped sample, the number of test values that were greater than or equal to a specified cut-off value were counted to provide the apparent prevalence for that bootstrapped sample and cut-off value. For each true prevalence proportion, 1000 bootstrapped samples were produced and the apparent prevalence for the specified cut-off value was recorded for each sample. The mean of the distribution of these apparent prevalence values provided a maximum likelihood estimate or expectation for the apparent prevalence at the specified cut-off value if a population with the specified true prevalence proportion was repeatedly sampled (Fig. 1).

To estimate the accuracy expected for a specific cut-off value, expectations for the apparent prevalence were determined for the cut-off value from the distributions of apparent prevalence from many (1000) bootstrapped samples across the complete range of prevalence proportions. Since the true prevalence was defined for each distribution by the proportion of sample values drawn from infected subjects, the expectation for the apparent prevalence was compared to the true prevalence to provide an estimate of the accuracy that would be expected across the complete range of prevalence proportions for the specific cut-off value.

The entire procedure was repeated for a range of selected cut-off values to estimate the accuracy expected for each cut-off value across the complete range of true prevalence proportions.

In our example, we produced bootstrapped samples each with 1000 values drawn from the samples of known infected and uninfected subjects to simulate a sample (n = 1000). The proportion of test values drawn from the sample of known infected subjects (true prevalence) within bootstrapped samples were: 0.01, 0.05, 0.10, 0.15, ... , 0.90, 0.95, 0.99. Cut-off values tested were 1.05, 1.10, … . 1.45, 1.50 PI. One thousand bootstrapped samples were produced for each cut-off value/true prevalence combination and the apparent prevalence for each bootstrapped sample was plotted in a distribution. For each cut-off value/true prevalence combination, the expected apparent prevalence was the arithmetic mean of the distribution and the 95% confidence limit values were the 2.5th and 97.5th percentiles of the distribution. Accuracy was estimated by subtracting the true prevalence from the expected apparent prevalence for each cut-off value/true prevalence proportion combination. Results were presented graphically. By visual inspection of the graphs, it was possible to select cut-off values that would be expected to perform optimally at various true prevalence proportions.

Two-Graph receiver operating characteristic (TG-ROC) cut-off value selection

An optimal cut-off value for the c-ELISA was estimated using TG-ROC analysis as previously reported (Greiner et al., 1995) on CMDT software (Jens Briesofsky, Institute for Parasitology and Tropical Medicine, Berlin Germany). Two-graph receiver operating characteristic analysis selects a cut-off value that maximizes sensitivity and specificity when sensitivity and specificity are

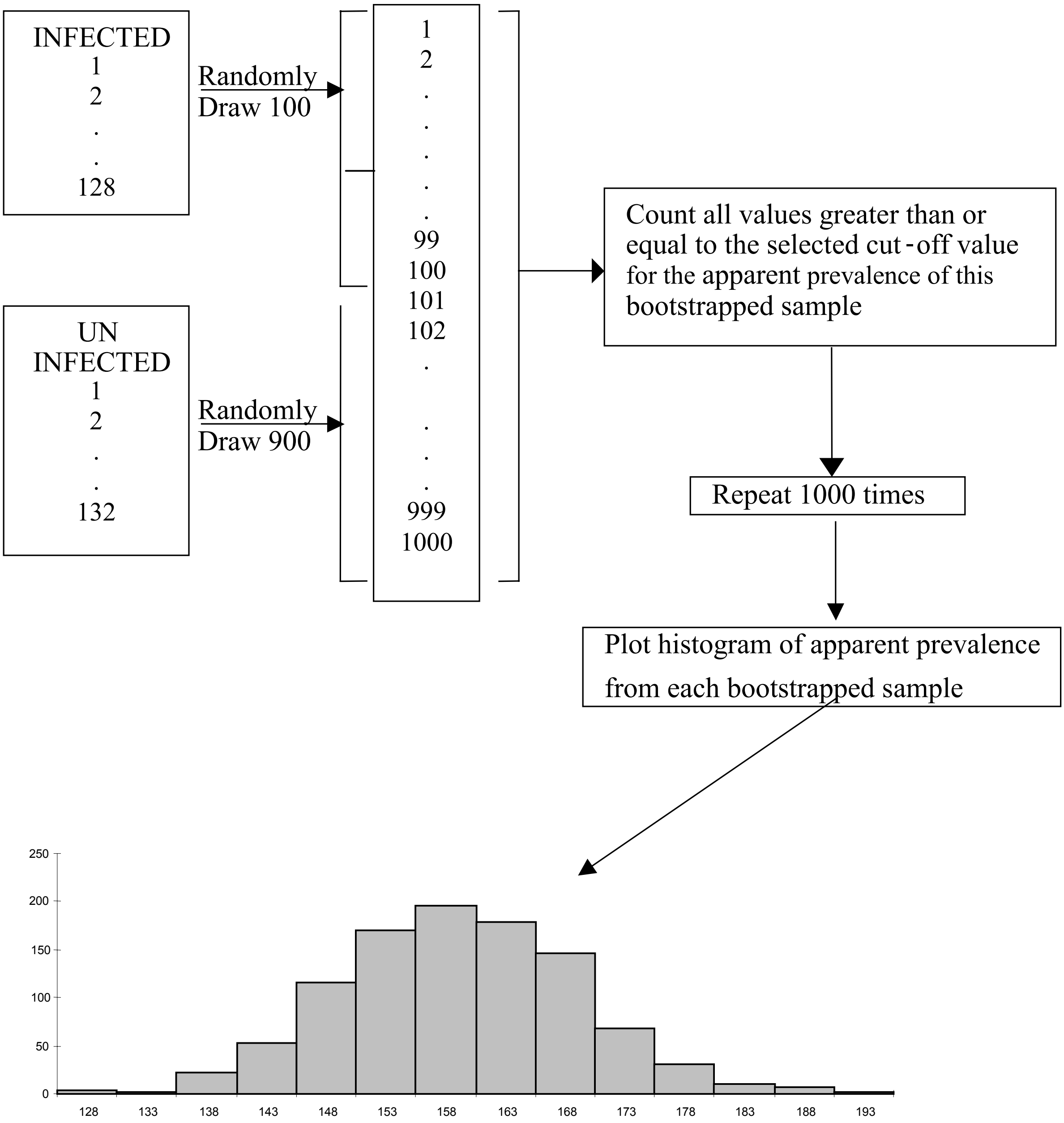


Fig.1 Schematic representation of bootstrapping to produce an expectation for the apparent prevalence of a sample (*n* = 1000) and a true prevalence of 10% for any selected cut-off value. The expectation for the apparent prevalence is the mean of the distribution and the 95% confidence limit values are the 2.5th and 97.5th percentiles of the distribution.

Bootstrapping/linear regression for estimating true prevalence

If the assumptions of the model hold for the population sampled and if the sample is a random or at least representative sample of the population, then the relationship between the true and expected apparent prevalence across the complete range of prevalence proportions for the cut-off value used in the survey can be used to estimate the true prevalence. For a sample of size *n*, bootstrapped samples can be produced each with *n* values randomly drawn from the samples of known infected and uninfected test values. The true prevalence in the bootstrapped samples can be varied across the complete range of prevalence proportions. Expectations for the apparent prevalence can be estimated for the cut-off value used in the survey across all true prevalence proportions. Since the relationship between true and apparent prevalence is linear across all prevalence proportions, a regression line and accompanying regression equation can be estimated by ordinary least squares regression for the cut-off value used in the survey. The regression equation can then be used to calculate the true prevalence from the apparent prevalence observed in the survey. Since the relationship between the true prevalence and the upper and lower 95% confidence limit values is also a linear relationship, the same procedure can be used to estimate a 95% confidence interval for the true prevalence.

To demonstrate the effect various cut-off values could have on true prevalence estimates, ordinary least squares regression were used to estimate regression equations for the apparent prevalence and accompanying confidence limit values for all cut-off values across the complete range of true prevalence proportions for a sample (n = 1000). We then produced apparent prevalence expectations for each cut-off value for samples (n = 1000) and a true prevalence of 0.20 by bootstrapping from the samples of known infected and uninfected test values. We used the regression equations for each cut-off value to estimate the true prevalence and confidence limit values from each apparent prevalence value. Results were presented graphically.

Rogan and Gladden's method for true prevalence estimation

Bootstrapping from samples of known infected and uninfected test values allowed the production of expectations for the apparent prevalence in samples of varying size and true prevalence using different cut-off values. The bootstrapped apparent prevalence expectations were used in Rogan and Gladden's equation (Rogan & Gladden, 1978; Greiner & Gardener, 2000b) to estimate the true prevalence and accompanying 95% confidence limits for all cut-off value/prevalence proportion combinations. Sensitivity and specificity values used in the equation were calculated for each cut-off value from the known infected and uninfected samples. Since the true prevalence was known, the error in Rogan and Gladden's estimates were expressed as the difference between Rogan and Gladden's estimate and the true prevalence. Results were presented graphically.

In order to assess the effect of different cut-off values on Rogan and Gladden's true prevalence estimates, bootstrapped apparent prevalence estimates for samples (n = 1000) and a true prevalence of 0.20 were corrected with Rogan and Gladden's formula for all cut-off values. Results were presented graphically and compared to true prevalence estimates produced with our bootstrapping regression method.

Bootstrapping software

All bootstrapped samples were produced with non-parametric bootstrapping using Resampling Stats® software (Resampling Stats Inc., Arlington, Virginia).

RESULTS

The relationships between bootstrapped expectations for apparent prevalence and true prevalence were linear for all cut-off values across most of the range of true prevalence proportions (Fig. 2). Deviations from linearity were slight and occurred only at extremely low and extremely high true prevalence proportions. Correlation coefficients were greater than 0.99 for all cut-off values and R-squared values for all regression lines were greater than 0.99.

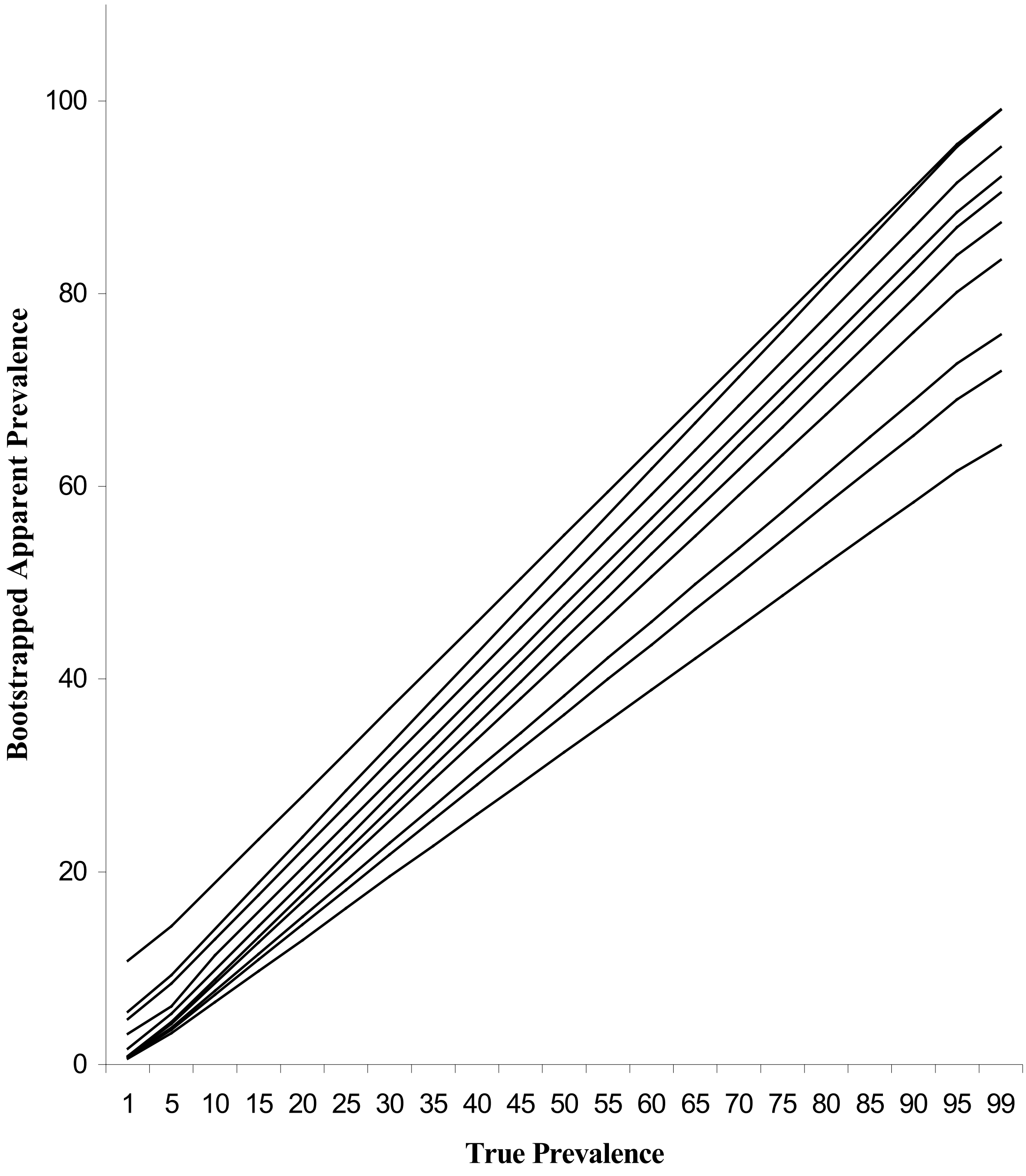


Fig. 2 Bootstrapped expectations for apparent prevalence versus true prevalence for each cut-off value. Each line represents a single cut-off value. Lines for cut-off values occur sequentially on the graph with the smallest cut-off value (1.05 PI) being the upper most line and the largest cut-off value (1.50 PI) being the lowest line.

The difference between bootstrapped expectations for the apparent prevalence and the true prevalence varied with both the cut-off value used and the true prevalence in the bootstrapped samples (Fig. 3). Cut-off values that were less than 1.10 PI overestimated the true prevalence. The amount of overestimation decreased as the prevalence increased. These cut-off values would be most accurate in samples with high true prevalence. Cut-off values that were greater than 1.30 PI underestimated the true prevalence. The amount of underestimation increased as the true prevalence increased. These cut-off values would be most accurate in samples with low true prevalence. Cut-off values between 1.10 and 1.30 PI overestimated true prevalence at low true prevalence proportions and underestimated true prevalence at high prevalence proportions. The cut-off value of 1.15 PI was the most accurate cut-off value across the complete range of prevalence proportions. It equally overestimated and underestimated true prevalence for true prevalence proportions below and above 50% respectively.

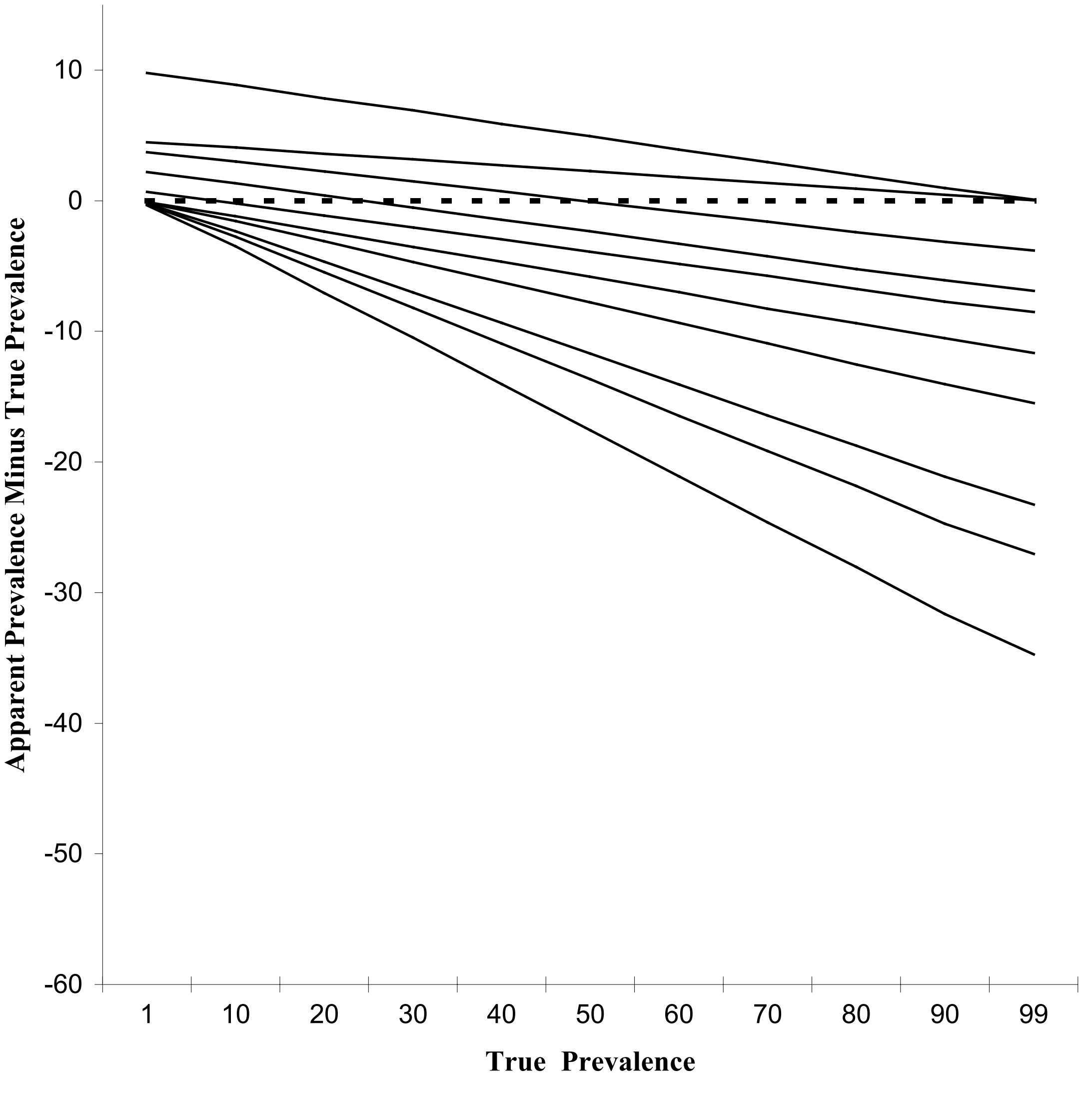


Fig. 3 Bootstrapped expectations for apparent prevalence minus true prevalence versus true prevalence for each cut-off value. Each line represents a single cut-off value. Lines for cut-off values occur sequentially on the graph with the smallest cut-off value (1.05 PI) being the upper most line and the largest cut-off value (1.50 PI) being the lowest. The dotted line is a reference line at zero difference.

Two-graph receiver operating characteristic analysis estimated the optimal cut-off value for the c-ELISA to be 1.15 PI (95% CI = 1.11 – 1.99). At this cut-off value, the sensitivity and specificity of the c-ELISA were estimated to be 96.15% (95% CI = 93.47 – 99.62).

Rogan and Gladden's equation overestimated true prevalence across the complete range of true prevalence proportions for cut-off values below 1.15 PI and underestimated the true prevalence for cut-off values above 1.15 PI (Fig. 4). There was complete agreement between the Rogan and Gladden estimates of prevalence and the true prevalence across all true prevalence proportions for the cut-off value of 1.15 PI.

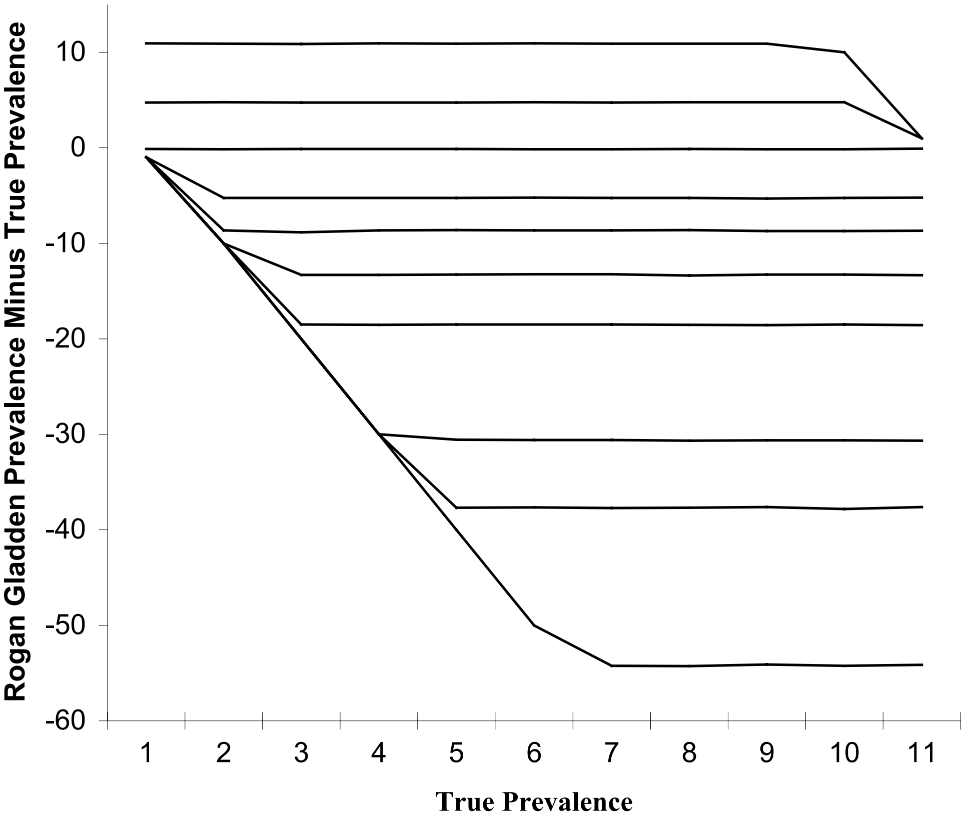


Fig. 4 Rogan Gladden true prevalence estimate minus true prevalence for each cut-off value. Each line represents a single cut-off value. Lines for cut-off values occur sequentially on the graph with the smallest cut-off value (1.05 PI) being the upper most line and the largest cut-off value (1.50 PI) being the lowest.

Figure 5 is a graphical presentation of the true prevalence estimated with Rogan and Gladden's method and our bootstrapping/linear regression method using all cut-off values for a sample (n = 1000) and a true prevalence of 20%. Bootstrapping expectations for apparent prevalence overestimated the true prevalence for small cut-off values. As the cut-off value was increased, the apparent prevalence expectations underestimated the true prevalence. Rogan and Gladden's estimates of true prevalence also varied from below 20% for small cut-off values to above 20% for large cut-off values. Rogan and Gladden's method produced true prevalence estimates that were more accurate than apparent prevalence estimates only for the narrow range of cut-off values around the optimal cut-off value of 1.15 PI (approximately 1.1 to 1.2 PI). Similarly, 95% confidence intervals for the Rogan and Gladden estimates of true prevalence encompassed 20% only for a narrow range of cut-off values (approximately 1.1 to 1.2). However, the bootstrapping/linear regression method provided estimates of true prevalence that were accurate across all cut-off values. Accompanying 95% confidence intervals also encompassed 20% for all cut-off values.

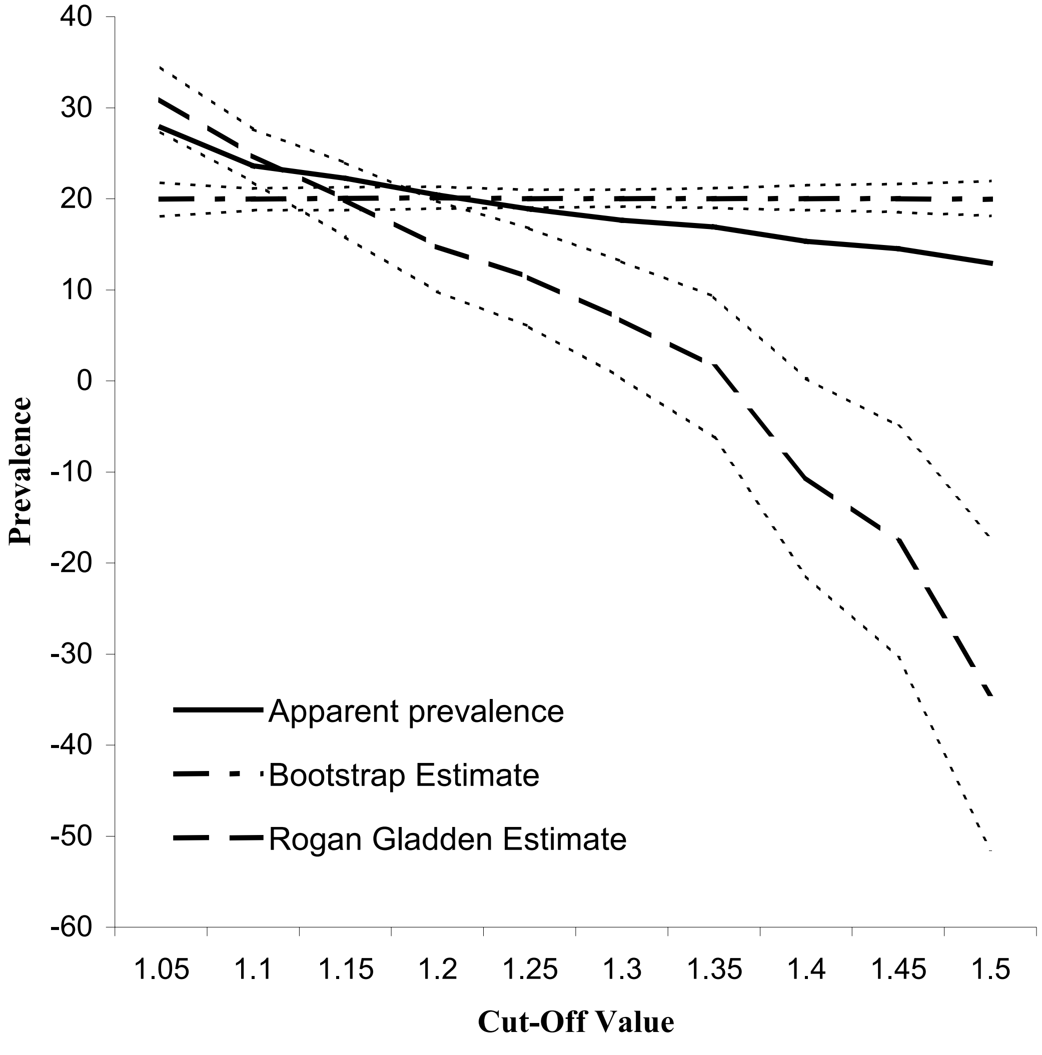


Fig. 5 Bootstrapping and Rogan Gladden estimates for true prevalence and accompanying 95% confidence bands when the true prevalence was 20% for each cut-off value. Apparent prevalence is the apparent prevalence expectation for a true prevalence of 20% for each cut-off value.

DISCUSSION

Bootstrapping can be used to produce unbiased estimates of population parameters from sample statistics (Efron & Tibshirani, 1993). If all assumptions were satisfied in our example, then the bootstrapped expectations for apparent prevalence could be considered unbiased estimates of the survey results that would be expected when testing bison populations with varying MCF virus infection prevalence using different cut-off values for the test. If the apparent prevalence estimates were unbiased, it should be valid to use them to evaluate the accuracy of different cut-off values and Rogan and Gladden's method. In addition, it should be valid to use the relationship between the apparent prevalence estimates and true prevalence to estimate the true prevalence in bison populations.

Although the relationship between the bootstrapped apparent prevalence and the true prevalence was linear for all cut-off values across the complete range of prevalence proportions (Fig. 2), none of the cut-off values were free from error across the complete range of true prevalence proportions (Fig. 3). Low cut-off values tended to overestimate the true prevalence and would be expected to be more accurate in samples with high true prevalence. Large cut-off values tended to underestimate the true prevalence and would be expected to be more accurate in samples with low true prevalence. The most accurate cut-off value was the optimal cut-off value as estimated by TG-ROC analysis. However, it correctly estimated the true prevalence only for a true prevalence of 50%. For all other true prevalence proportions it over or underestimated the true prevalence. Since no cut-off value was free from error and the true prevalence in population is unknown, all apparent prevalence estimates from surveys should be considered inaccurate and should be adjusted.

The accuracy of Rogan and Gladden's method for true prevalence estimation varied with the cut-off value selected and the true prevalence in the sample (Fig. 4). Cut-off values greater than the optimal value overestimated the true prevalence and cut-off values smaller than the optimal underestimated the true prevalence. The optimal cut-off value accurately estimated the true prevalence across the complete range of prevalence proportions. However, the accuracy of Rogan and Gladden's estimates were sensitive to even small variations from the optimal cut-off value. In the example of a population with a true prevalence of 20%, only a small range of cut-off values were more accurate than the apparent prevalence estimate. This was not the case for the bootstrapping/linear regression method, which accurately estimated the true prevalence regardless of the cut-off value selected.

The assumptions of this bootstrapping method for cut-off evaluation and true prevalence estimation are similar to those of previously reported methods, making this method prone to the same biases and weaknesses as other methods (Greiner & Gardener, 2000a; Greiner & Gardner, 2000b). Accurately determining the infection status of subjects for a validity study is often difficult (Greiner & Gardner, 2000a). For many infectious agents it requires testing with multiple reference tests or using other biological methods for ensuring that subjects are truly infected or uninfected (Greiner & Gardner, 2000a). In cases where it is not possible to accurately determine the infection status of subjects, other methods have been developed that allow the estimation of the sensitivity, specificity and prevalence from imperfect tests (Enoe et al., 2000). Assuming that the infected and uninfected subjects used in the validity study are representative of infected and uninfected subjects in the population being surveyed may not always be a correct assumption. Test values from uninfected subjects raised in isolation or in geographical areas free from the disease in question may be substantially different from test values of subjects in the population being surveyed (Greiner et al., 1994). For diseases in which there is an accurate reference test, randomly sampling and testing a portion of the survey sample can identify samples of infected and uninfected subjects upon which the bootstrapping method can be applied to estimate the true prevalence in the population.

The bootstrapping method presented in this report provides an alternative method for evaluating and selecting cut-off values as well as estimating the true prevalence in populations. The accuracy of bootstrapping prevalence estimates is much less sensitive to the cut-off value than Rogan and Gladden's method. However, both methods rely on similar assumptions and their estimates will be similarly biased when these assumptions are violated.

REFERENCES

Efron, B. and Tibshirani, R.J. (1993). An introduction to the bootstrap. Chapman and Hall, New York

Enoe, C., Georgiadis, M.P. and Johnson, W.O. (2000). Estimation of sensitivity and specificity of diagnostic tests and disease prevalence when the true diseases status is unknown. Prev. Vet. Med. 45, 61-81

Greiner, M., Franke, C.R., Bohning, D. and Schlattmann, P. (1994). Construction of an intrinsic cut-off value for the seroepidemiological study of Trypanosoma evansi infections in a canine population in Brazil: a new approach towards an unbiased estimate of prevalence. Acta Trop. 56, 97-109

Greiner, M. and Gardner, I.A. (2000a). Epidemiological issues in the validation of veterinary diagnostic tests. Prev. Vet. Med. 45, 3-22

Greiner, M. and Gardener, I.A. (2000b). Application of diagnostic tests in veterinary epidemiological studies. Prev. Vet. Med. 45, 43-59

Greiner, M., Sohr, D. and Gobel, P. (1995). A modified ROC analysis for the selection of cut-off values and the definition of intermediate results of serodiagnostic tests. J. Immunol. Meth. 185, 123-132

Li, H., Mcguire, T.C., Muller-Doblies, U.U. and Crawford, T.B. (2001). A simpler more sensitive competitive inhibition ELISA for detection of antibody to malignant catarrhal fever viruses. J. Vet. Diagn. Invest. 13, 361-364

Rogan, R.J. and Gladden, B. (1978). Estimating prevalence from the results of a screening test. Am. J. Epi. 107, 71-76

SPATIAL ANALYSIS OF BHV1 SEROLOGICAL STATUS IN PIEDMONT, ITALY, AS A GUIDE FOR DIFFERENTIAL ERADICATION STRATEGIES

N. VITALE*, L. CHIAVACCI, L. MASOERO, A. MENZANO, S. CALLEGARI AND A. MANNELLI

SUMMARY

A sample of 9706 animals, belonging to 392 cattle herds of the north western Italian region of Piedmont was tested for antibodies against BHV1 in 2002 – 2003. Geographic co-ordinates and information on potential risk factors for infection were collected. Overall prevalence of BHV1-positive herds was 69.4% (95% CI: 64.6 – 73.9). The spatial distribution of herds, based on serological status, was studied by a R – GRASS software interface. Exploratory kernel density estimates allowed the detection of a high prevalence area in the province of Cuneo, in southern Piedmont, where most of cattle farming is concentrated. Conversely, a low prevalence area was detected in the northern part of the province of Turin. Focussed K- function analysis was used to delimit the proposed low-BHV1-risk area, and significant clustering of BHV1 negative herds was shown between ~35 and ~58 km from a northern peak of kernel density. Accordingly, multiple logistic regression analysis resulted in a significant negative association between BHV1 positive status and proximity (within the first quartile of distance = 39.9 km) to the same kernel peak (OR= 0.46, 95% CI: 0.27 – 0.78). BHV1 positive test was positively associated with introduction of animals in a herd during the previous year (OR= 2.9, 1.7 – 5.0), and the natural logarithm of the number of animals in a herd (OR=3.0, 2.1 – 4.1). Based upon results of this study, differential eradication strategies could be suggested, taking into account the variations in BHV1 risk at the geographic level.

INTRODUCTION

Infectious bovine rhinotracheitis (IBR), caused by bovine herpesvirus type I (BHV1), causes economic losses due to abortion, infertility, respiratory disease, and low milk production (Hage et al., 1998). Moreover, trade restrictions for infected animals can be established in areas where the infection has been eradicated. Several countries within the EU have successfully eradicated BHV1 or have an EU-approved national voluntary or compulsory eradication programs. The strategies to control IBR are related to many factors, an eradication plan depends on level of seroprevalence, vaccination strategies, farming management. With high prevalence (> 50%) of infection the preferred strategy is vaccination and hygienic measures; while low prevalence (< 5%), especially if limited to old animals, is controlled with isolation and elimination of all seropositive animals. Therefore, before designing an eradication plan it is relevant investigating the BHV1 prevalence and related risk factors. Introduction of infected cattle and infected semen, and airborne transmission are recognised inter – herd BHV1 transmission routes (Van

---

* Nicoletta Vitale, Istituto Zooprofilattico Sperimentale del Piemonte, Liguria, e Valle d'Aosta, via Bologna, 148, 10154, Torino, Italy. E-mail: oss.epidemiologico@izsto.it

Wuyckhuise et al., 1998). Potential risk factors are herd size and farm density (Solis-Calderon et al., 2003). Geographic location should be considered in epidemiological studies because of heterogeneous spatial distribution of risk factors and potential viral transmission between neighbour herds. The identification of spatial variations in the risk of BHV1 infection, and the identification of areas characterised by different epidemiological status is critical for virus eradication strategies and allows better design of disease-control programs (McDermott et al., 1997; Ward & Carpenter, 2000).

Piedmont is one of the most important Italian regions for cattle farming, including ~ 18000 herds, 65% of which are concentrated in the plains between the provinces of Turin and Cuneo. About 33% of cattle belongs to the Piemontese breed, which is particularly appreciated for meat production. Economic viability of Piemontese breed farming can be hampered by IBR, mostly due to limitations to export. We carried out a serological survey on a sample of cattle herds in Piedmont in order to estimate BHV1 infection prevalence and to evaluate effects of herd – level risk factors. We used spatial analysis to study geographic variations in BHV1 risk. In fact, we hypothesized that the infection would be most prevalent in the previously mentioned high density area in southern Piedmont. Accordingly, the identification of areas characterised by a relatively low risk would allow the application of differential eradication strategies.

MATERIALS AND METHODS

Sampling and questionnaire design

The survey was conducted from February 2002 to January 2003. A proportional, stratified random sampling design was followed for herds. Stratification was carried out by health district, and herds size. Herd sample size was calculated to estimate an expected 50% prevalence with 5% precision, at the 95% confidence level (EPINFO, Version 6.04, Dean et al., 1994). Within each selected herd, individual animals were subsequently sampled. In small herds (<30 heads) all animal were sampled. In other herds, sample size was calculated to detect the infection if it was present in at least 10% of animals, with 95% confidence. The herd sampling frame was the Piedmont regional database for the identification and registration of cattle. A SAS® macro function was used to generate random numbers in sampling. Blood samples from selected animals were collected during mandatory testing against bovine brucellosis. The serum samples were tested for antibody against BHV1 glicoprotein gE by ELISA (IDEXX®). This test does not detect animals that were previously vaccinated with gE-deleted vaccines, whereas animals vaccinated with whole virus vaccines give positive results. Information on potential risk factors for BHV1-positive serological status at the herd level were recorder through a questionnaire that was administered to farmers by veterinarians at the time of blood sampling. There were 5 general questions about the farm (including the owner's address), 19 questions about herd management (including cattle-movement information, animals' breed, grazing pattern, breeding method, distance between herds, geographic elevation, health management, presence of other species) and 5 questions about IBR (clinical signs and vaccine strategies). Of the 29 questions included in the questionnaire, 10 were semi-open questions and 19 multiple-choice questions. Data were recordered into database (Microsoft ACCESS®).

Spatial analysis

UTM co-ordinates (zone 32) of farm locations were obtained using the Regional Technical Map (CTR), a raster map of Piedmont region, produced by the region's Cartography Service. A print out of such a map was sent by fax or e-mail to veterinarians of the local health districts who identified farm buildings on the map. Exploratory spatial analysis was carried by the R (www.r-project.org) GRASS (http://grass.itc.it) interface in the Mandrake 9.1 Linux operating system. A vector map of the region (www.regione.Piemonte.it) was imported in GRASS 5.0 and converted to raster format. The function *rast.get* was then used to import the map in the R software system. Two-dimensional kernel density estimation was carried out, using the *kde2d.G* function, for BHV1 positive and negative farms, and added to the raster as a contour map (Ihaka & Gentleman, 1996, Venables & Ripley, 1999).

Following exploratory analysis, and based on a *a priori* hypothesis of lower BHV1 risk in the northern part of Piedmont, we carried out focussed K-function analysis to demonstrate clustering of BHV1 negative herds around the point of maximum kernel density that we found for negative herds in northern Piedmont. K-functions were calculated for all farms, and for BHV1 negative farms, and subsequently compared (Fotheringham et al., 2000; Mannelli et al., 2003) by the following equation:

$$l(d) = \log(K_{BHV1-}(d) - \log(K_{allfarms}(d)$$

where $K_{BHV1-}(d)$ and $K_{allfarms}(d)$ were K-functions for BHV1− and all farms sampled, respectively. When $l(d)=0$, there was no difference in clustering between negative farms and all farms sampled. To test the null hypothesis that BHV1− were a random sample of all farms, we draw 95 random permutations of labels and plotted minimum and maximum simulated $l(d)$ as lower and upper simulation envelopes of observed $l(d)$. At distances where observed $l(d)$ was greater than maximum simulated values, we considered the observed result as significant at the 5% level (Diggle, 2003).

Analysis of risk factors from questionnaire data

Comparisons between BHV1 status of herds for each exposure level of categorical variables was carried out using the FREQ procedure in SAS® (SAS, 1999). Medians of number of animals per herd were compared by Wilcoxon two-sample test. Variables that were significant at the 15% level in bivariate analysis, were included in a multiple logistic regression analysis. Distance from the northern BHV1− focus was divided into quartiles and three dummy variables were coded in order to compare farms that were within the first quartile, and between first and second, and second and third, with those farms more distant that the third quartile as the baseline group. The likelihood ratio test was used to assess the overall significance of the model (two-tailed significance level $\alpha=0.05$) The significance of each term in the model was tested by Wald's χ^2. Estimated odds ratios (OR) and 95% Wald's CI were obtained as measures of predictor effect.

RESULTS

Serum samples from 9706 animals belonging to 392 herds were examined for antibodies against BHV1. Of the selected herds, 122 (31.1%) were dairy herds, 132 (33.6%) mixed herds and 138 (35.2%) beef herds. Because information collected on vaccination status of herds were considered as unreliable, all ELISA positive herds were classified as BHV1 positive, although test results might have been due to previous vaccination with whole virus vaccines (non gE-deleted). The herd – level BHV1 sero – prevalence was 69.4% (95% exact binomial CI: 64.6 – 73.9). The spatial distributions of BHV1+ and BHV1− herds are shown in Figures 1 and 2.

Although sampled herds were concentrated in southern Piedmont, between provinces of Torino and Cuneo, BHV1− farms show a second, northern cluster of kernel density corresponding to the UTM-32 co-ordinates X=414891, Y=5010417. According to comparison between focussed K-functions of BHV1− and all farms, BHV1− farms were significantly clustered around the above co-ordinates at distances comprised between ~35 and ~58 km (Fig. 3).

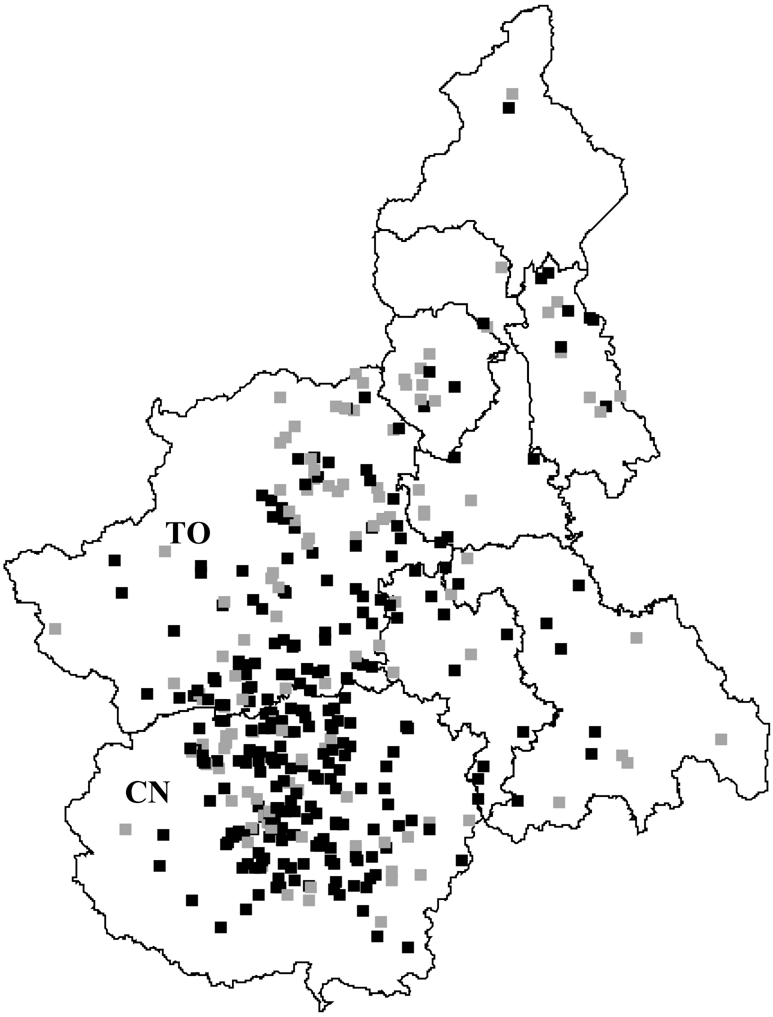


Fig. 1 Spatial distribution of farms that were examined for antibodies against BHV1 in Piedmont, in 2002-2003. Black and grey squares indicate sero-positive (BHV1+) and sero-negative (BHV1−) herds, respectively. TO: province of Turin, CN: province of Cuneo.

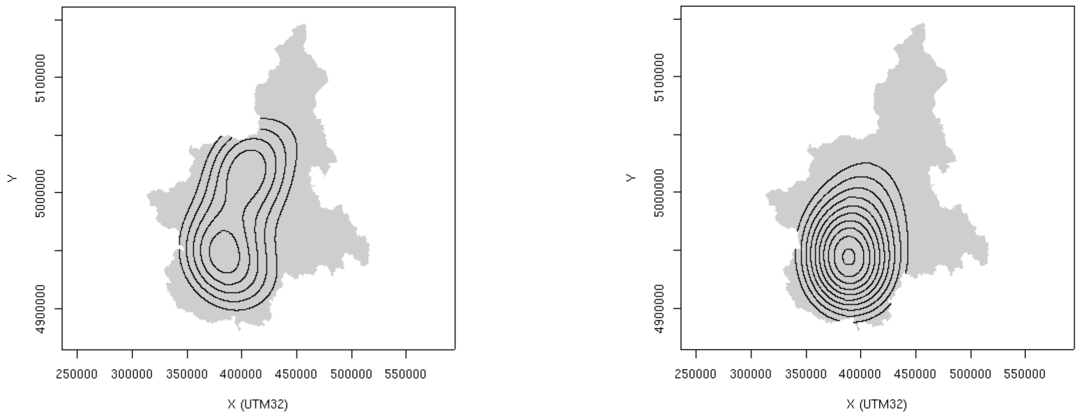


Fig. 2 Kernel density estimates of spatial distributions of a sample of cattle farms that were serologically negative (left) and positive (right) for antibodies against BHV1 in Piedmont, in 2002 –2003.

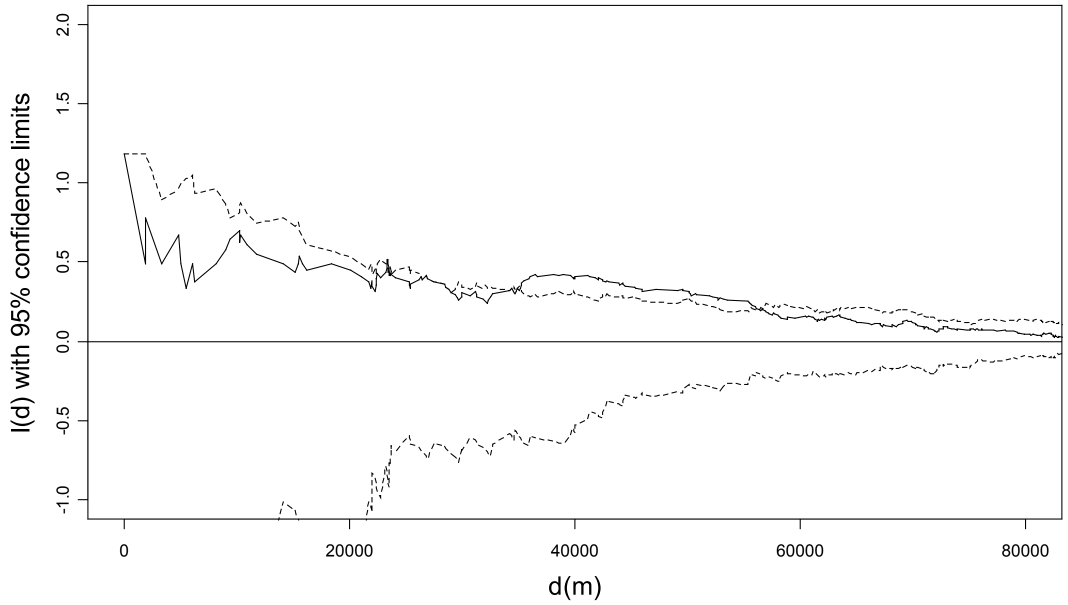


Fig. 3 Comparison of clustering of 120 BHV1− farms and 382 farms tested, around a BHV1− focus in northern Piedmont. The solid line represents the observed difference *l(d)* between K-functions, and dashed lines are lower 95% CI that were obtained by drawing 95 random permutations of labels.

Analysis of risk factors

Breed and most of farm management factors were not associated with BHV1 serological status. Fifty three out of 98 (54.1%) herds that were situated within the first quartile of distances (39.9 km) from the northern BHV1− focus were serologically positive, whereas prevalence in more distant herds was 74.5% (n=294). The final logistic regression model (likelihood ratio χ^2=87.0, d.f.=3, P<0.001, Table 1) included proximity to the BHV1− northern focus (distance < 39.9 km, Wald's χ^2=8.3, d.f.=1, P<0.01), introduction of animals in the previous 12 months (Wald's χ^2=15.1, d.f.=1, P<0.001), and the natural logarithm of the number of heads (Wald's χ^2=41.9, d.f.=1, P<0.001, Table 2). Hosmer and Lemeshow test showed no evidence of poor fit (χ^2=9.3, d.f.=8, P=0.32).

Table 1. Logistic regression analysis of risk factor for BHV1+ by ELISA in 392 cattle herds in Piedmont in 2002 – 2003.

RISK FACTOR	OR	95% CI
Proximity	0.46	0.27-0.78
Introduction of animals	2.9	1.7-5.0
Log number of heads	3.0	2.1-4.1

DISCUSSION

Using spatial analysis and multivariate logistic regression, we found spatial variations in the risk of BHV1 in Piedmont. Exploratory kernel density estimates and subsequent, focussed K-functions were used to delimit a low risk area in northern Piedmont, which can be considered as distinct from the main cattle production area in the south of the region. The combination of spatial analysis and logistic regression (that adjusted spatial effect for other farm – related predictors) allowed the identification of a ~ 40 km radius delimiting the low risk area. Factors underlying such spatial variations in BHV1 risk should be more thoroughly investigated, including inter herd viral transmission in high farm density areas. Nevertheless, based on results of this study, differential BHV1 eradication strategies could be suggested. In the high prevalence area, a gE negative, killed vaccine could be used to protect sero – negative herds from inter herd virus transmission, whereas gE negative, live vaccines could be considered for negative animals of infected herds. During the initial phase of eradication, positive animals could be vaccinated with whole virus, killed vaccines to limit viral excretion. A different policy could be suggested for the low risk area, where vaccination could be avoided in negative herds. In BHV1+ herds in the low prevalence area, slaughtering of positive animals could be considered, combined with vaccination of sero – negative animals with gE negative, killed vaccines.

ACKNOWLEDGEMENTS

The project was funded by Italian Ministry of Health, Department of Food, Nutrition and Veterinary Public Health. The project would not have been possible without the help of veterinarians of Piedmont health districts.

REFERENCES

Dean, A.G., Dean, S.A., Coulumbier, D., Brendel, K.A., Smith, D.C., Burton, A.H., Dicker, R.C., Sullivan, K., Fagan, R.F. and Arner, T.G. (1994). EpiInfo, Version Processing, Database and Statistics Program for Epidemiology on Microcomputer for Disease Control and Prevention, Atlanta, GA, 581pp

Diggle, P.J. (2003). Statistical analysis of spatial point patterns, Arnold. London, UK

Fotheringham, A., Brundson, C. and Charlton, M. (2000). Quantitative Geography. Perspectives on Spatial Data Analysis, Sage. London, UK

Hage, J.J., Schukken, Y.H., Digkstra, T.H., Barkema, H.W., Van Valkengoed, P.H.R. and Wentink, G.H. (1998). Milk production and reproduction during a subclinical bovine herpesvirus 1 infection on a dairy farm. Prev. Vet. Med. 34, 97–106

Ihaka, R., and Gentleman, R. (1996). R: a language for data analysis and graphics. J. Computat. Graph. Stat. 5, 299-314

Mannelli, A., Mandola, M.L., Pedri, P., Tripoli, M. and Nebbia, P. (2003). Association between dogs that were serologically positive for Rickettsia conorii relative to the residences of two human cases of Mediterranean spotted fever in Piemonte (Italy). Prev. Vet. Med. 60, 13–26

McDermott, J.J., Kadohira, M., O'Callaghan C.J. and Shoukri, M.M. (1997). A comparison of different models for assessing variation in the seroprevalence of infectious bovine rhinotracheitis by farm, area and district in Kenya. Prev. Vet. Med. 32, 219–234

SAS Institute Inc. (1999). SAS/STAT® User's Guide, Version 8, Cary, NC

Solis-Calderon, J.J., Segura-Correa, V.M., Segura-Correa J.C. and Alvarado-Islas, A. (2003). Seroprevalence of and risk factors for infectious bovine rhinotracheitis in beef cattle herds of Yucatan, Mexico. Prev. Vet. Med. 57, 199-208

Van Wuyckhuise, L., Bosch, J., Franken, P., Frankena K. and Elbers, A.R.W. (1998). Epidemiological characteristics of bovine herpesvirus-1 infections determined by bulk milk testing of all Dutch dairy herds. Vet. Rec. 142, 181–184

Venables, W.N., and Ripley, B.D. (1999). Modern applied statistics with S-Plus, Second Edition. Springer – Verlag, New York

Ward M.P. and Carpenter, T.E. (2000). Techniques for analysis of disease clustering in space and in time in veterinary epidemiology. Prev. Vet. Med. 45, 257–284

COMPANION ANIMALS

ILL HEALTH IN DOGS: DOES A TEMPORAL ASSOCIATION WITH VACCINATION EXIST?

D.S. EDWARDS*, W.E. HENLEY, E.R. ELY AND J.L.N. WOOD

SUMMARY

Some dog owners in the UK have raised concerns that vaccination may be associated with a wide number of health problems that affect dogs, ranging from minor conditions, such as bad breath and persistent scratching to serious illnesses, including epilepsy and cancer. One anti-vaccination campaign group have claimed that significantly more dogs suffer illness within three months of vaccination than dogs vaccinated more than three months previously. An epidemiological investigation was conducted to evaluate the evidence for a temporal association between vaccination and ill-health in dogs. Postal questionnaires were sent to the owners of a randomly selected sample of 9,055 dogs, 4,040 of which were returned. No temporal association was found between vaccination and ill-health in dogs after adjusting for potential confounders, including age, which was significantly associated with signs of disease. Using the Hauck-Anderson procedure, recently vaccinated dogs were found to have similar levels of ill health to dogs not recently vaccinated. Thus, within the acceptance limits of +/-5%, the recently vaccinated (<3months) and non-recently vaccinated groups had equivalent frequencies of disease.

INTRODUCTION

Vaccination has been a contentious issue since the introduction of the smallpox vaccine, with the ridicule and sensationalism in the 19th century press reflecting public anxiety and opposition (Bellaby, 2003). In recent years much media attention, including both television and newspaper coverage has been given to the claims of one anti-vaccination campaign group that one in ten dogs suffer illness within three months of vaccination, implying that canine vaccination is responsible for this ill-health due to impairment of the immune system (O'Driscoll, 1999; CHC, 2001). The anti-vaccination group based these claims on the results of a survey they conducted through a specialist dog magazine and on the analysis of signs of illness reported in dogs from a single veterinary practice during their first three-months post vaccination. They claimed that a wide range of conditions and signs of illness that affect dogs are caused by vaccination. Such a temporal association with vaccination, particularly if affecting 10% of vaccinated dogs, would be of great concern to dog owners and the veterinary profession. Conversely, a reduction of vaccine coverage in dogs could have serious implications not only for canine health but also for human health, in particular considering vaccination against canine leptospirosis.

---

* David Edwards, The Animal Health Trust, Lanwades Park, Kentford, Newmarket, Suffolk, CB8 7UU, UK. Email: david.edwards@aht.org.uk

A cross-sectional epidemiological study was conducted to assess the evidence for a temporal association between canine vaccination and ill health in dogs. Particular attention was paid to designing the questionnaire and selection of participants to reduce sources of bias. Simple equivalence testing methods were used to establish bounds on the size of any potential temporal association. The hypothesis tested was that recently vaccinated (<3 months) dogs do have a higher frequency of ill-health than dogs vaccinated more than three months previously.

MATERIALS AND METHODS

The study was designed to have 80% power to detect a two-fold increase in signs of ill-health for dogs vaccinated within the last three months, compared to dogs vaccinated prior to this. The required sample size was estimated to be 3,600 assuming an illness rate of 2% and 20% exposure. Exposure in this case was defined as dogs vaccinated within the previous three months.

British veterinary practices were contacted after being randomly selected from a national list. The study population consisted of dogs that had used the services of a veterinary practice within the previous twelve months. This included any transaction conducted in the dog's name and did not necessarily require the veterinarian to have seen the dog. Dogs were randomly selected from the computer databases of most practices that agreed to participate using MS Excel to generate a random selection of numbers within the range of the practice database. At one practice the dogs were randomly selected using the vaccine reminder list, restricting the dogs selected from this practice to animals vaccinated within the previous twelve months. Two practices had card-file rather than computerised records. A systematic sampling approach was used at one of these practices with the first number selected at random and every 20th card checked to see if the dog complied with the selection criteria. The other practice with a card-file system had all eligible dogs (135) included in the mailing due to its small size.

Pre-tested and validated postal questionnaires[1] were sent by the participating veterinary practices, each with a practice covering letter inviting the owner to participate in the study, a letter explaining the study and a reply paid envelope. One questionnaire was sent per household with the name of a specific dog to avoid selection bias by the owner in multi-dog households. The questionnaire, entitled 'Dog Health Survey' was carefully designed to avoid making the study hypothesis known to the respondent. Questions were asked regarding the dog's vaccination history and any signs of ill health shown by the dog within the two weeks preceding completion of the questionnaire. Details were asked about the dog, including any flea or worming treatments administered, and the dog's household. Reminder cards were sent after three weeks to owners who had not responded to the questionnaire. Details of the dog's last vaccination were checked with the dog's veterinarian, however with some practice records not every dog's vaccination dates could be checked. Demographic data were obtained from three of the participating practices for all selected dogs to compare between responders and non-responders.

Upon receipt completed questionnaires were checked for obvious errors and, where necessary, owners were contacted for clarification of responses. Data were recorded in a customised MS Access database, checked and examined using standard approaches for categorical data analysis (Agresti, 1990), in particular assessing associations between

---

[1] Questionnaire available from Animal Health Trust website: http://www.aht.org.uk/DHS.pdf

explanatory variables and signs of ill-health. A generalised additive modelling (GAM) approach, with cubic spline smoothers, was used to evaluate appropriate functional forms for the association between the outcome and continuous explanatory variables such as age and the number of vaccinations received (Hastie & Tibshirani, 1990). Multivariable logistic regression modelling was used to adjust estimates of the effect of recent vaccination on risk of ill-health for potential confounders. Apart from age, variables that did not confound the potential association between vaccination and ill-health were excluded from the models. The quality of model fit was assessed using the Hosmer-Lemeshow statistic (Hosmer & Lemeshow, 1989). Generalized logistic regression, a form of non-linear mixed effects modelling, was used to determine whether clustering of illness in dogs occurred at the practice or breed level (Gilmour et al., 1985). The Hauck-Anderson procedure was used to test for equivalence between the risks of ill-health amongst dogs vaccinated recently and those not vaccinated recently (Christley & Reid, 2003). The generalized logistic regression models were fitted using SAS/STAT with all other statistical computations performed using S-PLUS 6.1 (SAS/STAT, 2001; Insightful, 2001).

RESULTS

A total of forty-six veterinary practices were randomly selected from the national list of which 28 (61%) agreed to participate. From these practices 9,055 questionnaires were sent out with 3,966 useable questionnaires returned, representing an overall response rate of 44%. Demographic data were obtained for 667 non-responders' dogs.

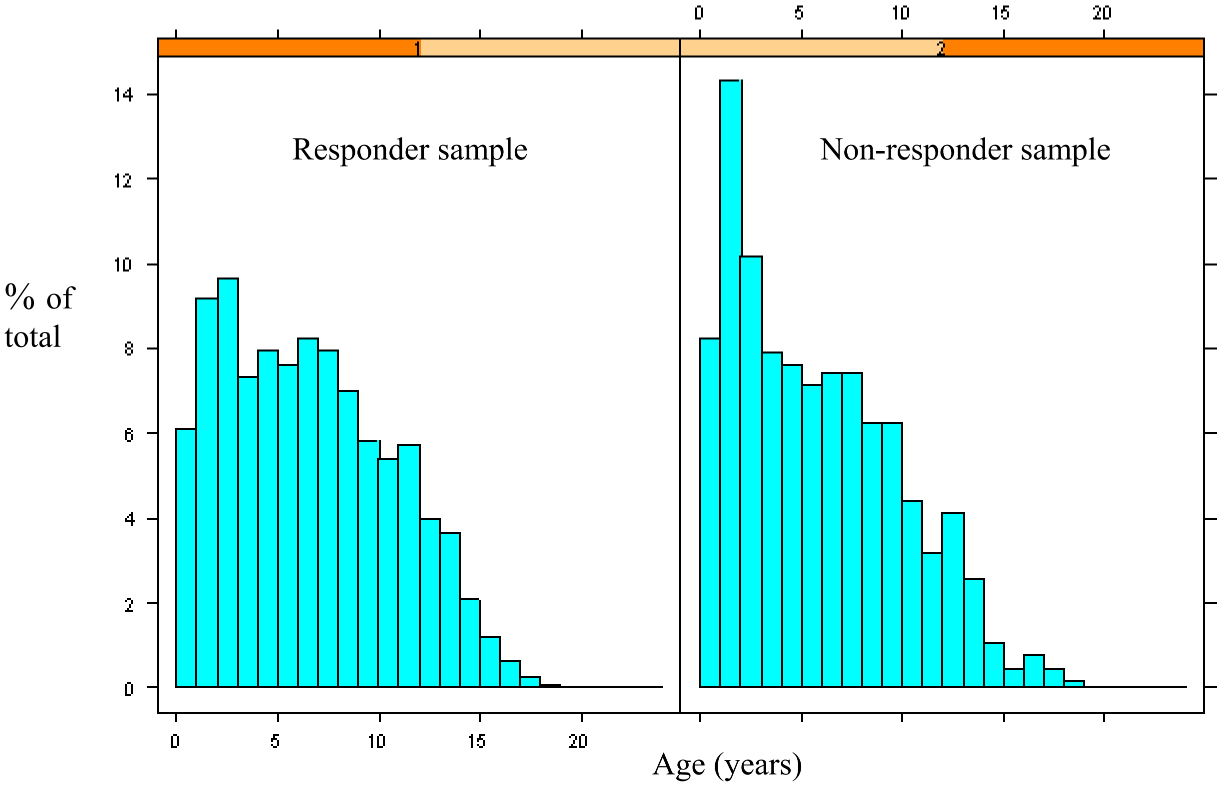


Fig. 1 Histograms comparing the age distribution of responders' dogs (shown on the left) and non-responders' dogs (shown on the right)

Dog gender was recorded as male in 51% of responses (2,002) and female in 49% of responses (1,943). Ages ranged from two weeks to 23 years (median 6 years 4 months). The age distribution for the 667 non-responders' dogs was similar, with an age range of three weeks to 20 years (median 6 years 3 months, Figure 1) and 57% of the non-responder's dogs were recorded as male. Figure 1 illustrates that the non-responder sample had a greater proportion of younger dogs than the main study group and with this exception the overall shape of the plots were similar (Figure 1). There were 124 breed groups represented with the most common being crossbred, Labrador Retrievers, Jack Russell Terriers, Border Collies and West Highland White Terriers (Table 1); again, the general distribution of dog breed within the responders and non-responders was similar.

Table 1. Most common breeds in the study dataset

BREED	NUMBER OF DOGS	PERCENTAGE OF SAMPLE	PERCENTAGE NON-RESPONDERS SAMPLE
		N= 3,966	N = 677
Crossbred	1,076	27.1	22
Labrador Retriever	348	8.8	8.4
Jack Russell Terrier	180	4.5	2.7
Border Collie	169	4.3	3.3
West Highland White Terrier	165	4.2	3.7
German Shepherd	162	4.1	4.8
Springer Spaniel	151	3.8	1.0
Yorkshire Terrier	150	3.8	3.9
Golden Retriever	144	3.6	4.0
Cocker Spaniel	116	2.9	1.0
Staffordshire Bull Terrier	101	2.5	4.9
Cavalier King Charles Spaniel	94	2.4	2.7
Poodle	76	1.9	2.5
Boxer	72	1.8	2.1
Dachshund	50	1.3	2.2
Collie	44	1.1	0.6
Other Breeds (107 breeds represented)	906	22.8	27

Nearly half of the dog households were situated in town centre or outskirts (1,892, 48%), followed by village or countryside (1490, 38%) and city centre or outskirts (520, 13%). Over one third of dogs were from multi dog households (1330, 34%) while approximately a quarter of dogs were from households that also had a cat (929, 24%). A quarter of the dogs were from households recorded as having some other type of pet (1,039, 26%). Ninety-six percent (3,810) of the dogs had received a wormer within the previous twelve months. Nearly four out of five (3124, 79%) of the dogs in the study had received flea treatment within the previous twelve months and one in four (1,012, 26%) of the dogs were from a household where the use of environmental flea treatment was reported.

Table 2. Commonly reported signs of ill health in study dogs and their association with recent vaccination

SIGN OF ILLNESS REPORTED BY DOG OWNER	NUMBER OF DOGS n=3,966	FREQUENCY PER 1,000 DOGS	ODDS RATIO [a] AND 95% CONF. INT.	P-VALUE [a]
Inflamed skin or persistent scratching	205	54.5	1.1 (0.8 – 1.5)	0.74
Stiffness	154	40.4	0.9 (0.6 – 1.3)	0.62
Limping	120	31.2	0.6 (0.3 – 0.9)	0.04
Weepy or inflamed eye(s)	115	30.0	0.4 (0.2 – 0.8)	0.02
Lump(s)	109	28.3	0.5 (0.3 – 1.0)	0.05
Sore/inflamed ear(s)	97	25.1	1.1 (0.6 – 1.7)	0.94
Diarrhoea	87	22.4	1.2 (0.7 – 1.9)	0.69
Bad breath	83	21.4	0.7 (0.4 – 1.3)	0.33
Vomiting	70	18.0	0.7 (0.4 – 1.4)	0.38
Hair loss	61	15.6	1.0 (0.5 – 1.8)	0.98
Persistent coughing	60	15.4	0.7 (0.4 – 1.4)	0.44
Eating less than usual	56	14.3	0.7 (0.3 – 1.4)	0.37
Tooth problems	49	12.5	1.0 (0.4 – 1.3)	0.33
Head shaking	48	12.2	0.9 (0.4 – 1.8)	0.79
Incontinence	45	11.5	1.2 (0.6 – 2.4)	0.78
Difficulty breathing	32	8.1	0.6 (0.2 – 1.8)	0.52
Fleas	32	8.1	0.7 (0.3 – 1.8)	0.56
Convulsions (fitting)	21	5.3	0.8 (0.3 – 2.5)	0.95
Blood in faeces/stools	19	4.8	0.5 (0.1 – 2.1)	0.49
Unable to stand or get-up	18	4.8	0.7 (0.2 – 2.5)	0.82
Unsteady	17	4.3	1.1 (0.4 – 3.6)	0.93
Abscess	15	3.8	1. 8 (0.5 – 6.1)	0.56
Nasal discharge	13	3.3	1.4 (0.4 – 4.5)	0.83
Ticks	14	3.5	0.6 (0.1 – 2.8)	0.78

a. Pearson's Chi-square

The number of vaccinations received by the dogs in the study ranged from zero to 24 (median 5). Time since last vaccination ranged from one day to 17 years (median 6 months). Twenty-three percent of dogs were recently vaccinated (<3 months), 1% (42) were unvaccinated and 1% (42) had their vaccination status recorded as unknown. The dogs' most recent vaccinations were predominantly combined boosters, including components to protect against distemper virus, para-influenza virus, canine parvovirus, *Leptospira interrogans* (serovars canicola and icterohaemorrhagica) and infectious hepatitis. Some dogs also had vaccinations administered against *Bordetella bronchiseptica* or against rabies. Over 18% of dogs had signs of ill-health within two weeks of questionnaire completion. The frequency of the most common signs of ill-health reported by dog owners is shown in Table 2. The percentage of ill-health amongst dogs recently vaccinated (<3months) was 16% compared with 19% of dogs not recently vaccinated (>3months), and was 25% in the dogs recorded as unvaccinated, or vaccination status unknown.

In univariable analyses, vaccination within the previous three months was not significantly associated with ill-health (Odds ratio [OR] 0.85, 95% confidence intervals [95%CI]: 0.69–1.03, P = 0.12). Recent vaccination was associated with reduced risk in the univariable analyses for three reported signs of illness, namely 'limping', the 'presence of a lump' and 'weepy or inflamed eyes' (Table 2). No statistical evidence of clustering at the veterinary practice or breed level was detected in the generalised logistic regression modelling (P=0.42 and P=0.89, respectively).

Logistic regression modelling indicated that signs of ill-health within two weeks of questionnaire completion significantly increased with age (OR = 1.11 for each year increase, 95% C.I. 1.09 – 1.13, P < 0.001). The GAM analysis found no significant non-linear variation in risk of ill-health with age (P = 0.14). After adjusting for age in the multivariable analyses, recent vaccination was not found to be significantly associated with ill-health (OR = 0.87, 95% C.I. 0.70 – 1.07, P = 0.18) (Table 3) and the model provided a reasonable fit to the data (Hosmer-Lemeshow statistic P = 0.12). Similarly, the number of prior vaccinations received was not associated with signs of ill-health in the dogs (OR=0.98, 95%CI: 0.95 – 1.00, P = 0.11) (Table 4) and provided a reasonable fit to the data (Hosmer-Lemeshow statistic P = 0.26). Neither the lack of significant association between recent vaccination and ill-health or the number of prior vaccinations received and ill-health was confounded by other variables, whether the variables were significantly associated with signs of ill health (e.g. age) or not (e.g. gender). In addition, no significant overall temporal associations were found between signs of ill-health and recent vaccination within preceding time intervals of 1 through to 12 months.

Table 3. Multivariable logistic regression model showing influence of recent vaccination (<3months) on frequency of reported signs of ill health in dogs

	COEF.	STANDARD ERROR	ODDS RATIO	95% C.I.	P-VALUE[a]
Intercept	-2.218	0.095			
Age (years)	0.107	0.011	1.11	1.09 – 1.14	< 0.001
Recent vaccination	-0.142	0.106	0.87	0.70 – 1.07	0.180

a. P-values calculated using likelihood ratio test

Table 4. Multivariable logistic regression model showing influence of number of vaccinations received on the frequency of reported signs of ill health in dogs

	COEF.	STANDARD ERROR	ODDS RATIO	95% C.I.	P-VALUE[a]
Intercept	-2.211	0.101			
Age (years)	0.121	0.014	1.13	1.10 – 1.16	< 0.001
Number of vaccinations	-0.023	0.014	0.98	0.95 – 1.00	0.105

a. P-values calculated using likelihood ratio test

Using the Hauck-Anderson procedure, recently vaccinated dogs had similar levels of ill-health to dogs not recently vaccinated (P=0.04 for the test of the null hypothesis that recent vaccination decreases risk of ill-health by at least 5% and P < 0.001 for the test of the null hypothesis that recent vaccination increases risk of ill-health by at least 5%). Thus, within the acceptance limits of +/- 5%, the recently vaccinated (<3months) and non-recently vaccinated groups were equivalent. The 90% confidence interval for the difference in risk of ill-health between vaccine groups indicated that recent vaccination does not increase signs of ill health by more than 0.1% and may actually be associated with a reduction of 4.9%.

DISCUSSION

The claim by anti-vaccination campaigners that one in ten dogs suffered illness within three months of vaccination has attracted media and public interest, potentially influencing future vaccine uptake. However, the claim was based on a limited review of one veterinary practice's records with no adequate control group for comparison, so that reported ill-health will have included chronic and other conditions unrelated to recent vaccination (CHC, 2001). Furthermore, the study by O'Driscoll (1999), that claimed significantly more dogs suffered signs of ill-health within three months of vaccination, was based on an unrepresentative magazine survey where the reader was aware of the hypothesis and no attempt was made to minimise responder or recall bias. Despite the limitations of the anti-vaccine group's magazine survey, the absence until now of suitable quantitative data has made it difficult to counter their claims. There are no reliable data that show rates of vaccination in the UK canine population, but a drop in rates of coverage could result in re-emergence in dogs of currently rare diseases such as canine distemper, and increased public health problems from canine zoonoses such as *L. interrogans* serovars canicola and icterohaemorrhagica, which are currently rare in both the canine and human populations (Ward et al., 2002). The diseases against which dogs in the UK are commonly vaccinated are now, with the exception of *Bordetella bronchiseptica*, thought to be rare.

This study was designed to provide quantitative data to test the hypothesis that ill-health is more frequent in recently vaccinated dogs. The sample size was chosen to provide sufficient power to detect a much smaller vaccination effect than that claimed by O' Driscoll (1999). When calculating sample size we were conservative in our estimate of the frequency of reported signs of ill-health, i.e. a calculation of 2% when the dataset had a frequency of over 18%. This was to ensure that the study had sufficient power to detect any association between recent vaccination and ill-health in dogs. When considering the apparently high frequency of ill-health reported by dog owners in this study it should be recognised that this includes minor signs such as bad breath, persistent scratching and eating less than usual (Table 2). This wide range of signs of ill-health, including minor signs, reflects the signs of illness that O'Driscol (1999) claimed to be temporally associated with vaccination in dogs. Also, this ensured that signs of illness that a dog owner may not consider bringing to the attention of their veterinary surgeon were included. However, this strength was also a weakness, in that the study was not restricted to veterinarian confirmed diagnoses. To ensure the validity of the study, dogs were selected for inclusion using two-stage cluster sampling and responders were blinded to the study hypothesis. Comparison of demographic data for the responders and a sample of non-responders suggested that the study population was representative in terms of age, gender and breed and did not indicate any systematic bias.

There was no comparison of health in vaccinated and unvaccinated animals because of the difficulty in identifying a comparable population of unvaccinated dogs: Veterinary practices only see small numbers of unvaccinated dogs and those that are seen are likely to be exposed to different environmental risk factors than vaccinated dogs, as the decision to vaccinate is likely, for example, to be influenced by the socio-economic status of the owner. Using other vaccinated dogs as controls and looking for a temporal association between vaccination and ill-health should have reduced the effect of unmeasured confounders.

Vaccination is not necessarily an innocuous procedure and adverse reactions to vaccines have been reported in the veterinary literature, usually anecdotally, generally being considered as rare events (Schultz, 1998). Recognised vaccine-induced conditions include anaphylactic reactions, autoimmune disease in genetically susceptible animals and possible post vaccine neurological disorders (Schultz, 1998). Most of the above conditions can be easily recognised as vaccine associated when they occur immediately or shortly after vaccination. The Suspected Adverse Reactions (SAR) reporting system in the UK is passive, depending on veterinary surgeons reporting any suspected reactions to the Veterinary Medicines Directorate (VMD) as well as to the vaccine manufacturer and therefore under reporting of vaccine reactions is likely. This makes the actual frequency of adverse vaccine reactions hard to quantify and provides uncertainty, which anti-vaccination groups can exploit by implying that serious complications are more frequent, helping to fuel public distrust in vaccines. A common result of successful vaccination campaigns, which lead to the apparent absence of the disease against which dogs need to be vaccinated against, is a questioning of the need for vaccination.

No evidence was found of a temporal association between recent vaccination and an increased frequency of signs of ill-health in dogs, either before or after adjusting for potential confounding variables such as age. This lack of association was consistent across the wide range of different time intervals used to define 'recent' vaccination, including from in-the previous month to the previous 12 months. Although the study was designed with adequate power to test the stated hypothesis, the absence of a statistically significant association did not necessarily imply that recent vaccination had no effect on canine ill-health. All that could be concluded was that there was an absence of evidence for a temporal association (Altman & Bland, 1995). We have found no evidence to support the claim for one in ten dogs suffering ill-health due to vaccination, as claimed by CHC (2001).

This study clearly indicated that older dogs have a significantly higher frequency of reported ill-health than younger dogs. Interestingly O'Driscol (1999) claimed that their survey did not support the view that as a dog gets older the frequency of illness will increase.

Assessing evidence for equivalence between the more recently and less recently vaccinated groups enabled important additional information to be gained. The Hauck-Anderson procedure was used to show equivalence of the two vaccination groups within the acceptance limits of +/-5%. In addition, by taking a confidence limits approach, the association was quantified more precisely. This analysis provided strong evidence against the claimed health risk associated with canine vaccination. Christley and Reid (2003) highlighted that although statistical methods for demonstrating equivalence are widely used in the pharmaceutical industry, it is common for medical and veterinary researchers to base assessment of equivalence on a failure to detect significant differences. Whilst this study was designed to test for temporal association rather than equivalence the use of such simple methods illustrates how they can be used to make more informative inferences about therapeutic equivalence for questions of current interest in biomedical research.

ACKNOWLEDGEMENTS

We thank the National Office of Animal Health for funding this research and the veterinarians and dog owners whose co-operation made this study feasible.

REFERENCES

Agresti, A. (1990). Categorical data analysis. John Wiley & Sons, New York

Altman, D.G. and Bland, J.M. (1995). Absence of evidence is not evidence of absence. Br. Med. J. 311, 485

Bellaby, P. (2003). Communication and miscommunication of risk: understanding UK parents' attitudes to combined MMR vaccination. Br. Med. J. 327, 725-728

CHC (2001). One in 10 risk of an adverse reaction from vaccines, New research from veterinary records. CHC Update 17, 1-3

Christley, R.M. and Reid, S.W.J. (2003). No significant difference: use of statistical methods for testing equivalence in clinical veterinary literature. J. Am. Vet. Med. Assoc. 222, 433-437

Gilmour, A.R., Anderson, R.D., and Rae, A.L. (1985). The Analysis of Binomial Data by Generalized Linear Mixed Model. Biometrika 72, 593 -599

Hastie, T. and Tibshirani, R. (1990). Generalized Additive Models. Chapman and Hall, London

Hosmer, D.W. and Lemeshow, S. (1989). Applied Logistic Regression. John Wiley, New York

Insightful (2001). S-PLUS 6 Guide to Statistics for Windows. Insightful Corporation, Seattle

O'Driscoll, C. (1999). What vets don't tell you about vaccines. Longnor, Derbyshire: Abbeywood Publishing

SAS/STAT (2001). SAS/STAT User's Guide, Version 8.2. SAS Institute Inc., NC

Schultz, R.D. (1998). Current and future canine and feline vaccination programs. Vet. Med. 203, 233-254

Ward, M.P., Glickman, L.T. and Guptill, L.E. (2002). Prevalence of and risk for leptospirosis among dogs in the United States and Canada: 677 cases (1970 – 1998). J. Am. Vet. Med. Assoc. 220, 53-58

TOWARDS REDUCING INJURY TO HORSE AND RIDER IN EVENTING: A CASE-CONTROL STUDY

J.K. MURRAY[1], E.R. SINGER, K.L. MORGAN, C.J. PROUDMAN AND N.P. FRENCH

SUMMARY

The objective of this study was to identify variables that increased or decreased the risk of a horse fall during the cross-country phase of an eventing competition. Data were collected for 180 cases and 540 controls. Cases were jumping efforts that resulted in a fall of the horse. Controls were matched by day and competition and were selected randomly from jumping efforts that did not result in a horse fall. The fence and event-related risk factors for horse falls in the final multivariable model were jumping into or out of water, taking off from good, good to soft or soft ground, fences with a drop landing, non-angled fences with a spread greater than or equal to two metres and angled fences. Other risk factors for horse falls were the rider's knowledge of their position before the cross-country, speed of approach, no refusals at earlier fences and cross-country lessons taken by the rider.

INTRODUCTION

Eventing is an equestrian sport enjoyed by amateur and professional riders, with 8106 riders registered with British Eventing in 2002. Five riders died in the United Kingdom (U.K.) as a result of horse and rider falls during the cross-country (XC) phase of eventing competitions during 1999. These fatalities raised concerns about the safety of this sport and initiated epidemiological investigations aimed at reducing the risk of injury to horse and rider.

Apart from these high profile deaths, the incidence rate for rider injury at event competitions has been reported as 0.88% (Paix, 1999). This rate is considerably higher than the injury incidence rates recorded by Chapman and Oni (1991) at an U.K. Grand Prix circuit for motorcycle racing (0.24%) and car racing (0.14%). Whitlock (1999) showed a rider injury incidence rate of 1.1% from data obtained from 54 days of XC competition in the U.K. including two fatalities, which resulted from the horse falling and landing on the rider. Previously, we reported that 32% of horses that fell were injured, with 1.5% being euthanased as a result of the injuries sustained (Murray et al., 2004). Horse falls occurring during the cross-country phase of events have thus been associated with injuries and fatalities to horses and riders.

Eventing consists of three stages: dressage, cross-country and show jumping. Events may be run over one, two or three days and are classified according to the level of difficulty of the cross-country course. There are 5 one-day event levels (in ascending order of difficulty): Intro,

---

[1] Jane Murray, Epidemiology Group, Faculty of Veterinary Science, University of Liverpool, Leahurst, Neston, Wirral, CH64 7TE, England

Pre-novice, Novice, Intermediate and Advanced. Three-day events are also knows as Concours Complet Internationale (CCI) competitions, with the difficulty increasing from 1-star (1*) to 4-star (4*) level. The stages of a one-day event are run in the order of dressage, show jumping and cross-country. Two-day events have dressage and show jumping on the first day and speed and endurance (incorporating the cross-country phase) on the second day. Three-day events have dressage on the first day, speed and endurance on the second day and show jumping on the final day. The speed and endurance day consists of four phases (phases A, B, C and D). Phase D is equivalent to the cross-country course at one-day events.

The cross-country stage (phase D) of three-day events covers a distance of 3900-7980 metres including a maximum of 30-45 jumping efforts over solid obstacles. Phase D requirements vary according to the level of the event (Anon., 2001). The cross-country phase of one-day events is shorter with course distances of 1600-4000 metres incorporating 18-40 jumping efforts. During the cross-country phase of eventing, the criteria on which horses and riders are judged include time, refusals, rider falls and horse falls. A rider fall incurs a penalty if the rider becomes separated from his/her horse in such a way to necessitate remounting. A horse fall is recorded if the shoulders and quarters of the horse touch either the ground or the obstacle and the ground at the same time (Anon., 2001). A horse fall, which incurs compulsory retirement, usually leads to a fall of the rider, and is therefore also known as a 'horse and rider fall'.

Analysis of retrospective data, using a case-control design, suggested an increased risk of falling associated with fences situated on a downhill slope (OR=8.4, 95% confidence interval (C.I.) =2.5-28.8, P=0.001) or fences with a ditch in front (OR=5.8, 95%C.I.=1.1-30.7, P=0.04) when compared with fences situated on flat ground and without a ditch in front (Singer et al., 2003). The risk of falling also rose as the number of fences on a course increased (OR=1.6, 95%C.I.=1.2-2.0, P<0.001). However, the risk associated with each additional jumping effort on the course decreased (OR=0.8, 95%C.I.=0.7-1.0, P=0.01).

In this paper, we report a large-scale prospective study conducted to investigate potential risk factors for eventing horse falls. The aims of the study were to identify variables that increased or decreased the risk of horse falls during the cross-country phase of eventing competitions. Ultimately, the risk factors will be reported to the sport's governing body so that intervention studies can be designed to reduce the risk of horse falls.

MATERIALS AND METHODS

Study design

A matched prospective case-control design with a ratio of 3 controls per case was used to test associations between event, fence, horse and rider related variables and horse falls. One- and two-day eventing competitions were randomly selected during the 2001 and 2002 British Eventing seasons. Only sixteen three-day event competitions were scheduled to take place during the study period and all sixteen were selected for inclusion so as to maximise data collection from three-day competitions. Data were obtained for 180 cases and 540 controls. Controls were individually matched by venue and day of cross-country competition, but not by class.

Case definition

A case was a jumping effort that resulted in a horse fall on the cross-country phase of an event. A jumping effort was defined as having occurred if the horse's front legs left the ground to commence take-off. A horse fall was defined as 'the horse's shoulders and quarters touched either the ground or the obstacle and the ground at the same time' which is the definition used within the British Eventing Rules (Anon., 2001). In this study, falls were identified and recorded by fence judges who were positioned by each fence to record penalties incurred by each competitor. The fence judges were briefed before the competition on the definition of a horse fall in order to maximise the probability of correct identification. Falls that occurred on the approach to the fence, as a result of the horse attempting to run out or refuse the fence, were not classified as cases.

Control definition and selection

A control was a jumping effort that did not result in a horse fall. Three controls were randomly selected from all successful jumping efforts that took place on the same day and at the same competition from which their case was selected. Matching was used to control for the potentially confounding effects of month, weather conditions and geographical location of the event.

Misclassification of the response variable was assessed by video analysis. Video footage was available for a total of 42 cases and 105 controls from three sources (BBC, Video Recall Ltd. and Lucid Dreams Ltd).

Data collection

Cross-country courses consist of between 16 and 42 numbered obstacles. The majority of obstacles consist of one fence, requiring a single jumping effort. However, some obstacles require multiple jumping efforts as they consist of two or more fences (also known as elements) situated in close proximity. These obstacles are known as combination fences. For the purposes of this study, two fences were defined as part of a combination fence if they were positioned so that the average horse would take four or fewer strides between the two fences. A four-stride distance between fences was equivalent to a mean (s.d.) distance of 16.8 (1.6) metres in this study.

For this study, every fence on the course was numbered consecutively and defined as a jumping effort. The order in which the riders started the cross-country course was used to identify in sequence, all jumping efforts that had been completed during the day. Thus, each jumping effort could be identified for the random selection of controls. In some situations, it was not possible to calculate the exact number of jumping efforts taken by a horse and rider combination, because competitors were given the choice between a shorter, technically difficult route, and an easier, but longer alternative with more elements. In situations where the exact number of jumping efforts was unknown, the minimum possible number of the jumping effort was used. Data were recorded on the day of the competition for fence- and event-level variables. The ground conditions were assessed subjectively by one of two observers who worked together at the beginning of the data collection period in an attempt to standardise interpretation of the five main categories of 'going' (firm, good-firm, good, good-soft, soft). The two observers also compared their assessment of ground conditions at events over the duration of the study so as to maintain consistency and reduce any effects of observer bias. On average, ground conditions were recorded at each fence every 3-4 hours. This enabled the conditions encountered by each horse in the study to be recorded as accurately as possible, by matching the time that each horse started the cross-country course to the nearest recorded ground conditions.

A fence was classified as having a drop landing if the ground level at landing was obviously lower than the ground level at take-off. Fences were also classified as being angled or non-angled. Angled fences were those positioned at an angle to the horse's line of approach. Angled fences may consist of a single or double set of rails (Fig. 1). Angled fences constructed from a single set of rails (Fig. 1A) require minimal width clearance from the horse, whereas 'corner' fences (Fig. 1B) require the horse to clear the width between both sets of rails in one jumping effort. The spread of the fence was defined as the distance (metres) that the horse would be required to clear, measured at the base of the fence. Fences were measured after the competition, so that hoofprints could be used to indicate where the majority of horses jumped the fence. The variables of fence angle and spread were shown to interact, so a new variable was created that combined the two variables. The new variable was assigned three categories (non-angled fence with a spread of <2 metres, non-angled fence with a spread of $\geq$2 metres and angled fences of any spread).

A: Angled fence

B: Angled fence (i.e. "Corner" fence)

Fig. 1 Schematic representation of the two main types of angled fences

A letter explaining the study was sent to all cases and controls within 3 days of the event. The letter informed the riders of imminent contact by telephone to complete a questionnaire relating to the event and additional areas such as horse and rider training. The telephone interviews were conducted as soon as possible with each rider selected as a case or control.

Risk of a horse fall

The risk of a horse fall was calculated by dividing the number of horse falls by the total number of jumping efforts and was expressed as the risk per 1000 jumping efforts. The risk was calculated as a function of the level of difficulty of the cross-country course and the type of event (one-, two- or three-day event).

Data analysis

All variables were tested for association with falling using univariate conditional logistic regression models. The statistical packages R (www.r-project.org) and Egret® (Cytel Software Corporation, USA) were used for data analysis. Continuous variables were also categorised into quintiles in the univariable analysis. The fit of the categorical variables in the model were compared to the fit of the continuous variables by assessing the change in deviance, (assuming the change in deviance follows a chi-squared distribution with n degrees of freedom, where n is the number of extra parameters fitted). To reduce the effects of collinearity, continuous variables were centred by subtracting the mean of the variable from all recorded observations (Kleinbaum et al., 1988).

Variables with a P-value <0.2 were considered for inclusion in a multivariable model, which was built using the technique of backward elimination. The variables ‘total number of jumps’, ‘jumping efforts on the course’, ‘ditch in front’, ‘downhill approach’ and ‘jumping effort number’ were also considered for inclusion in the multivariable model as a result of *a priori* evidence of an association identified by Singer et al. (2003). The effect of biologically plausible interactions between variables was also tested for in the model. Rider status (professional or amateur event rider), event type (one- or three-day event) and level of the event (Intro/pre-novice, novice, intermediate or advanced) were evaluated as potential confounders. A change in the regression parameters of $\geq$25% was considered to be indicative of confounding. The fit of the model was assessed by examination of the sensitivity and specificity of the model at cut-off points ranging from 0.2 to 0.6. Model stability was assessed by examination of the delta betas. The model was considered to be stable if removal of individual cases or controls altered the odds ratio by <25% and did not affect the significance of individual variables.

Analysis of potential recall bias

In order to investigate the potential presence of recall bias, the dressage score reported by each rider during the telephone interview was compared with that officially recorded at the event. Recall accuracy was classified as a binary variable (accurate=0, inaccurate=1) rather than as a continuous variable as the data were not normally distributed and could not be transformed to a normal distribution. Dressage scores were awarded in penalty points, therefore the lower the penalty point score, the better the performance. One-day event dressage tests are marked by a single judge and therefore result in penalty scores that are integers. Three judges are used to mark three-day event dressage tests. The average mark awarded by the judges for a three-day event competitor, rounded to two decimal places, forms their dressage score. To allow for differences in reporting style between riders (integers or decimals), a recalled score was considered accurate if it was within 0.5 penalty points of the British Eventing database score.

The effect of case-control status of the respondent and time between questionnaire and event was investigated by including these as explanatory variables in generalised linear mixed models. Case-control status and time since the event were introduced as fixed effects into the model. The potential confounding effects of rider status (professional or amateur event rider) and event type (one- or three-day event) were also considered. Generalised linear mixed models were fitted to the data using the function, glmmPQL, in the statistical package R, with the matching variable as the random effect (Armitage et al., 2002). Variables with $P \leq 0.05$ were considered to be statistically significant.

RESULTS

The overall risk of a horse fall was 0.35 falls per 1000 jumping efforts (95%C.I.=0.30-0.41). The risk appeared to increase as the level of difficulty and duration of the event increased (Table 1). Three-day events and one-day event championships were noted to be associated with a higher risk of a horse falling compared to one-day events.

All the variables considered for inclusion in the multivariable model, as a result of *a priori* evidence of an association identified by Singer et al. (2003) were excluded from the final model, as their inclusion did not improve the fit of the model (P>0.05). These were ‘a downhill approach to the fence' (P=0.42), 'ditch in front' (P=0.10), 'jumping effort number' (P=0.97), 'total number of jumping efforts on the course' (P=0.19) and 'total number of obstacles on the course' (P=0.25). The final model is shown in Table 2. No interaction was found between variables in the model and there was no evidence of confounding by the variables of rider status, event type and event level.

Table 1. A summary of the number of falls per 1000 jumping efforts recorded at randomly selected eventing competitions in Great Britain during 2001-2002.

Class	No. of falls	No. of jumping efforts	No. of falls per 1000 jumping efforts (95% C.I.)
One-day events			
Intro	2	18988	0.11 (0.00 - 0.38)
Pre-novice	20	131026	0.15 (0.00 - 0.24)
Novice	42	178106	0.24 (0.17 - 0.32)
Intermediate	37	68044	0.54 (0.38 - 0.75)
Advanced	14	24841	0.56 (0.31 - 0.95)
[a]CIC 1-star	1	6383	0.16 (0.00 - 0.87)
[a]CIC 2-star	1	9529	0.10 (0.00 - 0.58)
[a]CIC 3-star	0	4537	0.00 (0.00 - 0.81)
Novice Championship	2	1373	1.45 (0.18 - 5.24)
Intermed. Championship	2	1778	1.12 (0.14 - 4.05)
One-day Event Total	121	444605	0.27 (0.23 - 0.33)
Novice-two day event	1	2847	0.35 (0.00 - 1.96)
Three-day events			
[b]CCI 1-star	16	24049	0.66 (0.38 - 1.08)
[b]CCI 2-star	15	14950	1.00 (0.56 - 1.65)
[b]CCI 3-star	9	12346	0.73 (0.33 - 1.38)
[b]CCI 4-star	18	10563	1.70 (1.01 - 2.69)
Three-day Event Total	59	61908	0.95 (0.73 - 1.23)
Overall total	180	509360	0.35 (0.30 - 0.41)

[a] CIC = Concours Internationale Combined, (International one-day event)

[b] CCI= Concours Complet Internationale, (International three-day event)

Table 2. Multivariable conditional logistic regression model of risk factors for cross-country horse falls at eventing competitions in Great Britain (2001-2002).

Variable	Coefficient	Standard Error	Odds Ratio	95% C.I	LRT P-Value
Take-off surface					
Firm / Good-firm	Ref.		1.00		
Good	0.83	0.38	2.29	1.09 – 4.82	
Good-soft / Soft	2.74	0.93	15.56	2.54 – 95.45	
Water	3.91	0.80	49.80	10.38 - 238.99	<0.001
Landing surface					
Firm / Good-firm	Ref.		1.00		
Good	-0.47	0.40	0.63	0.28 - 1.38	
Good-soft / Soft	-0.51	0.94	0.60	0.09 - 3.83	
Water	1.74	0.51	5.72	2.12 - 15.45	<0.001
Landing					
Without a drop	Ref.		1.00		
Drop landing	1.23	0.38	3.41	1.60 - 7.25	0.001
Angle and spread of fence					
No angle, <2m spread	Ref.		1.00		
No angle, ≥2m spread	1.18	0.33	3.24	1.71 - 6.16	
All angled fences	1.57	0.43	4.83	2.09 - 11.16	<0.001
Position before XC					
Didn’t know position	Ref.		1.00		
First	1.48	0.57	4.39	1.44 - 13.43	
Second or lower	-0.81	0.35	0.45	0.22 - 0.89	0.001
Approach speed					
Appropriate	Ref.		1.00		
Too slow	1.61	0.49	5.00	1.90 - 13.14	
Too fast	1.84	0.44	6.30	2.64 - 15.02	<0.001
Previous XC refusals on the course incurred by the horse and rider					
Earlier refusals	Ref.		1.00		
No refusals	3.14	1.04	23.02	2.98 - 178.12	0.003
Rider has XC lessons					
No	Ref.		1.00		
Yes	0.66	0.24	1.94	1.21 - 3.09	0.006

The fit of the model was assessed by examination of the delta betas. Individual cases and controls with delta betas greater than 0.2 and less than –0.2 were removed from the dataset and the model was rerun. The variables of ‘drop landing’, ‘angle and spread of the fence’, ‘cross-country lessons’, ‘approach speed’ and ‘previous refusals on the course’ were considered to be stable, as the odds ratios altered by <25% and the significance of the variables remained unchanged. The variables of ‘take-off surface’, ‘landing surface’ and ‘position prior to the cross-country’ were less stable as the individual removal of 8 cases and 10 controls altered the odds ratio by >25%, although the interpretation of the significance of the variables did not change in relation to the critical P-value of 0.05. Examination of these cases and controls revealed no unusual covariate patterns and the individuals were therefore left in the dataset. The predictive capacity of the model was assessed by calculating the sensitivity and specificity of the model at various cut-off points (Table 3). The specificity of the model was shown to be good, although the sensitivity was poorer, particularly at cut-off values of 0.4 and above. Thus, the model was able to predict controls better than cases when the cut-off value was 0.4 or higher.

Table 3. Sensitivity and specificity of the multivariable conditional logistic regression model shown in Table 2 at cut-off points 0.2 to 0.6.

Cut-off point	Sensitivity (% of cases predicted)	Specificity (% of controls predicted)
0.2	86.7	74.1
0.3	74.1	84.4
0.4	68.1	88.1
0.5	59.0	93.0
0.6	47.6	96.5

Misclassification

Accuracy of the classification of the response variable was assessed by studying video footage that was available for 42 cases and 105 controls. Classification was 100% accurate for controls and 83% (35/42) accurate for cases. Five of the misclassified cases were horses that had stumbled badly and fallen onto their knees, but not fulfilled the case definition that required the shoulders and quarters to touch the ground. The other two cases that had been misclassified were ‘rider only’ falls, caused by the horse hitting the fence hard. However, neither of these two horses fell onto their knees, shoulders or quarters.

Recall bias

Univariable analysis showed a significant relationship between the number of days since the event and the accuracy of dressage score recall ($P<0.0001$). Dressage scores were less likely to be reported accurately as the number of days increased between the event and questionnaire completion [regression parameter β (SE) = 0.05 (0.01)]. The case-control status of the rider ($P=0.48$) was not associated with the accuracy of the score reported by the rider. There was no evidence of confounding between the variables investigated.

DISCUSSION

This study has identified a number of risk factors for cross-country horse falls. Surface type at the approach of the fence was significantly associated with an increased risk of horse falls. Fences jumped out of water were associated with a high risk of a horse fall, compared to fences with a firm or good to firm take-off ($P<0.001$). Fences requiring a take-off out of water were usually sited so that horses would take at least one stride in water before jumping. The increased risk of falling at fences with a take-off in water may have been attributable to miscalculation of the jump height and take-off point by the horse and rider due to the base of the fence being obscured under water, and / or the drag of the water unbalancing the horse. This finding along with evidence from an earlier study, suggest that water fences should be considered as an important area of risk on cross-country courses. Our earlier findings (Murray et al., 2004) showed an increased risk of injury for falls in water compared to falls on land (OR=2.1, 95%C.I.=1.1-3.7, P=0.01). Whilst jumping into water is considered by some to be an important test of the horse's 'bravery' and obedience, jumping out of water is less of a test for the horse. This was reflected in the higher proportion of refusals recorded at fences jumped into water (10.15 per 1000 jumping efforts) as opposed to fences jumped out of water (0.45 per 1000 jumping efforts), (Murray et al., unpublished data). Fewer horse falls would be anticipated if competitors exited water complexes by cantering up a slope rather than by jumping out of the water.

Good take-off ground (OR=2.3) and good to soft or soft take-off ground (OR=15.6) were associated with a higher risk of falling when compared to firm and good to firm take-off ground. Human long jump and high jump athletes use a firm take-off surface to aid performance and land on a soft surface to help minimise the risk of injury (Fukuda, 1988). It is possible that equine jumping performance might also be enhanced and the risk of falling reduced, by the provision of good to firm take-off surfaces at cross-country fences.

The width of the fence and the angle of approach were identified as significant risk factors for horse falls. The increased risk of a fall associated with non-angled fences having a base spread of two metres or more, compared with those with a spread of less than two metres, may be due to insufficient impulsion needed to clear the height and spread of the fence. No relationship was observed between the risk of falling and the spread of the fence once the base spread reached two metres or more (P=0.5). During the study period, the maximum permitted base spread was 2.8 metres for a fence that required vertical clearance and 3.5 metres for an open ditch (Anon., 2001). These results suggest that reducing the maximum permitted base spread for fences may reduce the risk of a fall.

Angled fences include corner fences and those positioned in such a way that horses were required to jump the fence at an angle. These fences are considered to test the horse's obedience, as penalties may be incurred easily at these fences, as a result of the horse running past the fence. A logical explanation for the increased risk of falling associated with angled fences is that the horse needs to make additional adjustments at take-off to ensure that both front legs are raised sufficiently in order to clear an angled fence.

The association between fences with drop landings (i.e. the ground level at landing was lower than the ground level at take-off) and an increased risk of a horse fall (OR=3.4) is a logical association that may be explained by a loss of proprioceptive balance of the horse on landing. In contrast with the findings of Singer et al. (2003) who found that fences sited downhill were associated with an increased risk of a horse fall (OR=8.41, 95%C.I.=2.46-28.78, P=0.001), our results showed no significant association between fences with a downhill approach and the risk of a horse fall (OR=0.93, 95%C.I.=0.56-1.53, P=0.76). The conflicting findings attributed to fences with a downhill approach may be as a result of the different methods of data collection. Singer et al. (2003) collected data retrospectively, which may have led to inaccuracies in the recall of the gradient of the approach to the fence and misclassification of some cases. Additionally, Singer et al. (2003) found fences with a ditch in front to be associated with an increased risk of a horse fall (OR=5.77, 95%C.I.=1.09-30.68, P=0.04). Although our study showed no significant association between fences with a ditch in front and the risk of a horse fall (OR=0.65, 95%C.I.=0.31-1.36, P=0.25), the confidence intervals from the two studies do overlap. The contradictory findings may be partly explained by the different case definitions that were used in the two studies. Singer et al. (2003) included falls that resulted from a refusal. These falls were excluded from our study because the selection of controls did not include attempted jumping efforts that resulted in refusals. Our study excluded six falls that occurred as a result of a refusal, three of which were at fences with a ditch in front. Falls following a refusal may be more likely to occur at fences that have a ditch in front, since the forward momentum of the horse can carry it into the ditch, thus causing a fall. The differing case definitions may therefore explain the different conclusions drawn as to the risk associated with fences with a ditch in front.

The rider's knowledge of their position within the competition at the start of the cross-country phase was associated with the risk of a horse fall. Riders who knew that they were in the lead prior to starting the cross-country course were at a higher risk of falling (OR=4.4) than riders who were unaware of their position. In contrast, riders who knew that they were not in the lead were at a lower risk of falling (OR=0.4) than those that were unaware of their position. It could be hypothesised that horse and rider partnerships that achieved excellent dressage scores (and were therefore in first place) were less proficient at jumping cross-country fences than partnerships that achieved less good dressage scores, thus placing them at an increased risk of a horse fall. Alternatively, this finding may be explained by the fact that riders who knew that they were currently in first position may have been more likely to 'take a risk' during the cross-country phase, in the hope of maintaining their lead in the competition. Conversely, riders who were not in the lead prior to the start of the cross-country may have been riding more cautiously or 'safely' with the priority of completing a round without jumping penalties, irrespective of the time taken.

Riders who had not incurred any refusals on the course, prior to selection, were at an increased risk of a horse fall compared with riders that had already been penalised for a refusal. An explanation for this finding is that some horses and riders would rather attempt to jump a fence and risk a fall than incur a refusal, despite sometimes reaching the fence at a poor take-off point or with inappropriate speed, balance or impulsion. Competitive and determined riders may be very reluctant to allow their horses to refuse. If their horses try to refuse a fence, then they may ride strongly, perhaps using their whip and spurs to encourage their horses to jump. An obedient horse may then attempt to jump the fence, and possibly fall, rather than risk punishment for having refused. Conversely, some horses rarely refuse and can be seen to jump at speed with little apparent care for their own safety. The temperament of these horses leads them to attempt to clear a fence and risk an awkward jump or fall, in preference to refusing.

Riders who believed that they had approached the selected fence at an appropriate speed had a lower risk of falling than riders who reported an approach speed that was too slow (OR=5.0) or too fast (OR=6.3). This finding is particularly interesting as inappropriate speed was anecdotally reported to be a contributory factor to some of the fatal falls that occurred during 1999. However, this result should be interpreted with caution as the competitor's retrospective opinion of their speed may be subject to reporting bias. Cases may have been more likely than controls to report an inappropriate speed, in an attempt to find an explanation for their fall. Subjective verification of the speed of a sample of cases and controls needs to be conducted to assess whether bias is present.

Our study found that those riders who received cross-country tuition had an increased risk of falling (OR=1.9) compared with riders that did not receive cross-country tuition. An explanation may be that those riders that took cross-country lessons were doing so because they were aware that they needed tuition to improve their performance on the cross-country course. It is interesting to note that of the riders selected for this study, only 46% (322/694) received cross-country lessons, whilst 94% (651/694) of riders received dressage tuition and 86%

(594/694) of riders received show jumping lessons. Further research is recommended to explore the effects of increasing the proportion of riders receiving cross-country tuition in relation to the risk of a horse fall.

In a previous study, we found that the risk of falling increased as the number of fences on a cross-country course increased (Singer et al., 2003). As a result of the matching used in the present study, the cases selected from classes with the greatest number of fences (CCI 3-star and CCI 4-star) were matched to controls competing over the same courses, as these were the only classes held at their respective venues. This could explain the apparent lack of association between number of fences on a course and the risk of falling found in this study.

There was no difference between cases and controls in the accuracy of reporting dressage scores by telephone interview. However, there was evidence of an effect of the number of days between the event and telephone interview. The most plausible explanation for this is memory decay in the reporting of dressage scores. Although it is possible that as the respondents were forewarned of the telephone interview, they may have kept a copy of their results near the phone for a few days in preparation for the interview. Memory decay was not confounded by case-control status. Our findings highlight the importance of minimising the time period between the event and questionnaire completion in retrospective data collection. However, the time delay experienced in our questionnaire completion was largely due to the non-availability or evasiveness of some of our respondents, rather than insufficient time being allocated for interviewers, and was therefore difficult to improve.

Video analysis indicated some misclassification of cases (7/42) by fence judges. It is expected that the risk factors associated with the stumbles experienced by five of the misclassified case horses would be similar to the risk factors identified for horse falls by this study. The potential effect of the misclassification of cases is currently being investigated but is not expected to be large.

This study has identified a number of risk factors for horse falls that are associated with the fence and the rider. We are currently in discussion with the sport’s officials as to how our findings on course design might be tested through suitable intervention studies.

ACKNOWLEDGEMENTS

The study was funded by the Home of Rest for Horses. The authors would like to thank all those riders who completed questionnaires for this study, fence judges for identifying and recording cases and the staff at British Eventing for providing access to their database. Additionally, the authors are most grateful to Jane Barnes for her assistance with telephone questionnaires and data entry. We are also grateful to the Federation Equestre Internationale (F.E.I.) for funding the pilot retrospective study.

REFERENCES

Armitage, P., Berry, G. and Matthews, J.N.S. (2002). Statistical methods in medical research, Fourth edition. Blackwell Science, London, UK.

Anon., (2001). British Eventing Rule Book. British Eventing, Warwickshire.

Chapman, M.A.S. and Oni, J. (1991). Motor racing accidents at Brands Hatch, 1988/9. Br. J. Sports Med. 25, 121-123

Fukuda, H. (1988). Biomechanical analysis of landing surfaces with different stiffnesses. Biomechanics XI-B, 679-684. Free University Press, Amsterdam.

Kleinbaum, D.G., Kupper, L.L. and Muller, K.E. (1988). Applied regression analysis and other multivariable methods. Second edition. PWS-KENT Publishing Company.

Murray, J.K., French, N.P., Saxby, F. and Singer, E.R. (2004). A retrospective study of horse falls occurring during the cross-country phase of horse trials. Vet. Rec., In press.

Paix, B.R. (1999). Rider injury rates and emergency medical services at equestrian events. Br. J. Sports Med. 33, 46-48

Singer, E.R., Saxby, F. and French, N.P. (2003). A retrospective case-control study of horse falls in the sport of horse trials and three-day eventing. Equine Vet. J. 35(2), 139-145

Whitlock, M.R. (1999). Injuries to riders in the cross-country phase of eventing: the importance of protective equipment. Br. J. Sports Med. 33, 212-216

POLICY

AN ASSESSMENT OF THE ANNUAL LIKELIHOOD OF THE INTRODUCTION OF FOOT AND MOUTH DISEASE VIRUS AND CLASSICAL SWINE FEVER VIRUS TO THE DANISH PIG POPULATION

B.M.DEC. BRONSVOORT[1], L. ALBAN AND M. GREINER

SUMMARY

Denmark is the largest exporter of pork world-wide with 85% of production going for export. Therefore, the consequences of any introduction of foot and mouth disease (FMD) and classical swine fever (CSF) would be enormous. This paper deals with the first part of a risk assessment on the likelihood of foot and mouth disease virus (FMDV) and classical swine fever virus (CSFV) introduction into Denmark.

The methodology involved the development of scenarios based on material from scientific articles, reports from veterinary agencies, and through consultation with experts. Specific information on the quantities of animals and commodities both legally and illegally traded was obtained from the Customs Officers and Veterinary Services and included both Danish and other experiences.

The preliminary results indicate a number of pathways or hazards for the introduction of FMDV and CSFV to Denmark. These include airborne spread, movements of animals or their genetic material, returning livestock trucks, wildlife, hunters, the trade in animal products and illegal imports of animal products from outside the EU. This assessment has identified airborne spread, trade in genetic material and returning livestock trucks as particular hazards for the introduction of FMDV. Climatic models are in place and used whenever there is an outbreak of FMD in a neighbouring country. The trade in boar semen, returning trucks and illegal meat imports are the most important pathways for CSFV. Much of the threat comes from trade with Germany due to CSF in the wild boar population there. Denmark has lowered the risk associated with returning trucks by implementing a mandatory washing, disinfection and 48-hour ban on trucks entering livestock premises. In addition, the amounts of illegally imported meat are small. The assessment has also identified a number of areas where the information is currently insufficient to make reliable judgements. For example, the possible risks of hunters returning with infected wild boar meat from other EU and third countries, or the carriage of infected meat from Eastern European countries by migrant farm workers are not clear.

The risk assessment process has resulted in an evaluation of the current regulations in the Veterinary Services and the Danish swine industry. It is planned to develop an adaptable framework based on the current results that can be updated easily as new threats arise. This is

---

[1] Mark Bronsvoort, University of Edinburgh, Centre for Tropical Veterinary Medicine, Royal (Dick) School of Veterinary Studies, Easter Bush, Roslin, Midlothian, EH25 9RG, UK.

important as risks will change as patterns of trade and demography alter. Such assessments should be carried out in 'peace time' when a more rational approach can be adopted and information more easily obtained.

INTRODUCTION

Despite its limited size, Denmark has a substantial swine industry. Almost 24 million slaughter pigs were produced in Denmark in 2002. The Danish pork industry is worth over 3 billion Euro per year. In total the Danish swine industry makes up approximately 9% of the Danish gross domestic product. Denmark is the 9th largest pig producer in the world and the largest exporter of pork in the world, with an annual export of over 1.6 million tonnes, accounting for around 85% of the national production. The USA and Canada are the second-largest exporters of pork. Approximately 60% of Danish export is to the EU market, mostly the to UK and Germany with the main third country being Japan. Given the size and importance of the export market, it is critical to the Danish swine industry to maintain the national swine herd free of diseases that could cause even temporary suspension of trade. The Office International des Epizooties (OIE) include foot-and-mouth disease (FMD), classical swine fever (CSF), swine vesicular disease and African swine fever in the List A classification of diseases that affect swine. Denmark, as a member of the World Trade Organisation, is bound by the Sanitary and Phytosanitary agreement and therefore must base its sanitary measures on international standards where these exist.

Table 1. Number of outbreaks of List A swine diseases in Europe between 1999-2001 (source OIE Handistatus).

Disease	Country	No. of outbreaks 1999-2001
FMD	France	2
	Greece	14
	Ireland	1
	Netherlands	27
	United Kingdom	2,034
CSF	Austria	1
	Germany	969
	Italy	17
	Luxembourg	7
	Spain	33
	Switzerland	49
	United Kingdom	16

Historically, Europe has experienced outbreaks of FMD and CSF with these have continued in recent years (Table 1). This situation is likely to continue and may even worsen with the forthcoming enlargement of the EU. One of the main problems with making risk assessments for disease introductions is the lack of reliable information on the pathways and the volume of movement along particular pathways. There are many potential pathways and some proven routes, such as illegal animal movements or infected trucks for FMD and contact with wild boar for CSF. However, in many cases the source of the original introduction cannot be verified definitively, making it difficult to be sure of the importance of many routes. For example, between 1990 and 1999, following the ban on vaccination against FMD in Europe, there were

30 introductions into Europe. Of these, six were attributed to purchases of animals, seven were attributed to fomites, five to spread by humans, one listed as other and 11 unknown (de Vos et al., 2000).

The likelihood of introduction of foot and mouth disease virus (FMDV) or classical swine fever virus (CSFV) by a pathway will depend on the prevalence of disease in the country of origin, the trade contacts, movements of animals and people and survival of the virus in the environment. Access to various sources of data and information is a vital requirement for risk assessments of this kind. The study has used the national research network of the International EpiLab in Denmark to meet this requirement (Greiner, 2003). There is often considerable uncertainty about the probability of any pathway, however, the relative probabilities for different pathways, products or countries may help in planning the allocation of resources for mitigative measures. The process of carrying out a risk assessment as thoroughly as possible helps to identify areas where reliable information is lacking, or where previously an activity was not considered a risk but in the light of new trade patterns, should now be reconsidered.

The aim of this project was to identify disease introductory pathways or hazards that could result in the introduction of FMDV or CSFV to the swine industry in Denmark. Having identified the pathways, the next aim was to assess the volume of trade or scale of movement of a product in order to make a qualitative assessment of the likelihood of disease introduction via this pathway. Ultimately, the aim is to build this information into an adaptable framework where the likelihoods and overall risks can be recalculated easily as new information becomes available on changing patterns of trade.

MATERIALS AND METHODS

This assessment incorporated a number of components including a literature review, discussions with experts in a wide range of fields and eliciting specific information regarding volumes of movements of animals, trucks and people from the relevant Danish government agency. This information was used to develop the pathways specifically for the Danish situation and to quantify the movements along each branch. In the process, new or more appropriate pathways were developed, which required new information resulting in an iterative process. These pathways then formed the basis for the qualitative assessment.

In addition, 25 of the Danish government field veterinarians were asked to participate in a simple ranking exercise. These veterinarians were attending a training course and were used as a convenient sample of government veterinary opinion. They were asked to rank a list of disease introductory routes for FMD and CSF, and to rank the regions of the world according to the likelihood that an outbreak in the next five years would originate there. The mean, mode and median ranks for each were then estimated.

RESULTS AND DISCUSSION

Veterinarians: The ranking of the importance of the various pathways by Danish Veterinary Service veterinarians is summarised in Table 2. Only the FMDV data are presented here because of space limitations. According to these veterinarians, illegal meat imports and airborne spread are the most important routes of introduction into Denmark, and Eastern Europe and Eastern Asia (including China) were considered the regions from which FMD is most likely to originate. This compares with two studies of expert opinion which both concluded that illegal import of livestock and livestock products were the most import, and that airborne spread as a route of primary introduction was the least important route (Horst et al., 1996; Gallagher et al., 2002).

Table 2. Danish Veterinary Service veterinarians' ranking of likely routes of introduction of FMDV into Denmark in the next five years (1 most likely to 12 least likely) ordered highest to lowest based on the mean ranking.

	Ranking		
Route of FMDV introduction into Denmark	Mean	Mode	Median
Illegal meat or meat products	3.8	1	4
Airborne spread	4.2	1	4
Illegal import of infected live animals	4.4	5	5
Illegal import of animal products other than meat	4.8	4	4
Tourist/immigrant foodstuff	4.8	4	4
Contaminated returning livestock vehicles	5.5	5	5
Carried by wild animals or birds into Denmark	5.7	6	6
Legal import of infected live animals	6	6	6
Illegal swill feeding kitchen or catering waste	6	5	5
Legal meat or meat products	7.1	6	7
Legal import of animal products other than meat	7.2	8	7
Imported bedding	8.2	12	10

Airborne spread: The last outbreak of FMD in Denmark in 1982 was most likely due to airborne spread from infected pig farms in the northern part of the former East Germany. This area suffered from a contemporary outbreak caused by the same virus strain. The disease appeared to spread across the Baltic Sea to the islands of Funen and Zealand, where cattle became infected (Westergaard, 1982). A number of models are available for predicting high risk areas for spread (Donaldson et al., 2001). The presence of large swine units in northern Germany, the high cattle densities in Denmark, and the possibility of introduction of FMDV into Germany from Eastern Europe mean that this route remains an important pathway for Denmark, although with a low probability. Very little can be done to reduce the risk from this route, although climatic models developed by the Danish Meteorological Institute can be used to identify high risk areas where surveillance may need to be increased (Sørensen et al., 2001).

Live Animal Movements: Clearly the potential for the introduction of FMDV via live animal movements should never be underestimated as large amounts of virus may be present in the secretions and excretions of infected animals. In the 2001 FMD epidemic, the disease was spread to France and The Netherlands through legal movements of sheep that were incubating the disease, and transmitted the virus to a consignment of calves at a truck stop in France. From Table 3 however, it is clear that Denmark imports very few cloven-hooved animals. The main imports are of cattle from The Netherlands, pigs from France and sheep and goats from Spain. Although regulations permit free movement of animals within the EU, the swine industry operates a voluntary quarantine of animals following import, while the cattle industry has a mandatory quarantine in the country of origin. This 30-day holding period allows disease to be detected before the animals enter Danish farms. Therefore, the probability of importing either CSFV or FMDV from these countries via live animal imports is low (Horst et al., 1998). The likelihood of the introduction of CSFV is possibly higher as CSF may be subclinical and not detected during quarantine.

Table 3. Number of live animal imports into Denmark in 2002 by country of origin and species.

Species	Exporting country	Total (n = 771)
Cattle	France	21
	Netherlands	190
	Italy	1
	Sweden	90
	Germany	19
	Austria	4
Pigs	France	203
	Germany	1
	Sweden	1
	UK	50
Sheep and goats	Netherlands	64
	Spain	122
	Germany	5

Returning Livestock Trucks: In contrast to live animal imports, Denmark exports millions of animals, mainly pigs, to the rest of Europe with 88% of this trade being with Germany. The numbers of recorded border crossings by trucks is given in Table 4. A previous study on returning trucks suggested that those returning from pig farms were not acceptably clean (Rugbjerg et al., 1998). At ambient temperatures FMDV (Bachrach et al., 1957) and CSFV (Edwards, 2000) may survive for several weeks in the environment. Since 2001, all returning trucks are required to have been cleaned thoroughly prior to crossing into Denmark (in accordance with EU directive and extended with Danish order no. 773) and are then required to be disinfected at an officially approved station in Denmark. Drivers are issued with a certificate and are then obliged to wait a further 48 hours before the truck can enter livestock premises where a veterinarian must be present to collect the papers proving that the truck was disinfected. Because of its geographical position, Denmark can monitor its borders relatively easily. In addition, the disinfection procedures are now well established, and compliance is believed to be very high according to the people involved in monitoring animal transport. The incorporation of a well-managed cleaning programme at the border and 48-hour ban on entering livestock premises should reduce the likelihood of disease entry to negligible and low levels for FMDV and CSFV, respectively.

Table 4. Numbers of truck movements entering and leaving Denmark each year by species and product carried.

Trucks transporting	Year		
	2000	2001	2002
Pigs (out of Denmark)	4,802	4,505	5,575
Pig feed (into Denmark)	119	1,510	4,280
Cattle (out of Denmark)	881	875	995
Cattle feed (into Denmark)	497	1,152	1,118

Genetic Material: Embryos and semen are now widely traded around the world. They are of particular concern from a disease transmission viewpoint, as virus can be directly implanted into a susceptible animal. This pathway has been well known for bovine embryos and the OIE guidelines, which include washing the zona pellucida to remove any viruses, reduce the risk to posed to a negligible level (Sutmoller & Wrathall, 1997). Denmark imports only bovine embryos and the annual numbers of imports are very small, for example the total imports for 2002 were only 1,487 embryos (Table 5). In addition, the imports are all from countries recognised by the OIE as free of FMD and therefore the probability of introducing FMDV via this route is negligible.

Table 5. Importation of bovine embryos to Denmark 2002.

Country	No. batches	No. embryos
Austria	12	127
Canada	27	215
France	44	239
Germany	32	207
Italy	20	120
Netherlands	84	404
Sweden	5	50
Luxembourg	1	7
USA	28	118
Total	253	1,487

On the other hand, Denmark imports large numbers of bovine semen doses (Table 6) and a smaller number of porcine doses (Table 7). Bovine semen straws are held for 30 days after collection while the donor is monitored for signs of clinical disease, which considerably reduces the likelihood of introduction, as clinical signs of FMD should become apparent in cattle within 14 days of infection. Although the number of imported bovine semen doses is large, they originate from countries recognised by the OIE as FMD-free. Therefore, the probability of importing FMDV via bovine semen imports is considered low to negligible.

Table 6. Importation of bovine semen in 2001 (source Annual Report 2001, Dansk Kvægavl)

Country of origin	No. of semen doses imported
USA	9,554
Canada	18,789
Netherlands	5,940
France	1,841
Germany	13,484
Sweden	18,170
Italy	2,933
Spain	360
Austria	1,589
England	440
Finland	1,000
Total	74,100

The officially reported annual numbers of porcine semen doses imported is relatively small at just under 4,000 (Table 7). However, there is some doubt about this figure as this information appears not to be recorded centrally, although it should appear on the Animal Movement (ANIMO) database. Unlike bovine semen, porcine semen is generally used fresh, and therefore boars need to be quarantined before collection, and also examined then to confirm absence of clinical signs of FMD and CSF. CSF is of much more concern via this pathway since the clinical signs are often very mild and the incubation period can be protracted. Although there is no record of disease introduction to a country via this route, the CSF outbreak in The Netherlands in 1997 involved two boar stations, and infected semen was believed to have been responsible for the infection of up to 36 farms (Hennecken et al., 2000). All boars on an AI station are tested for CSFV on entry and departure. The two Dutch boar stations are believed to have been infected by the introduction of boars infected during transport on a contaminated lorry (Elbers et al., 1999). This highlights the need for a quarantine period as well as the need for the proper cleaning of trucks. Denmark imports semen mostly from The Netherlands and Germany, where there are regular CSF outbreaks, although good surveillance and control measures are in place in these countries. Therefore, there is a low probability of importing CSFV via boar semen, although there needs to be further investigation of the porcine semen trade.

Table 7. Number of boar semen doses imported into Denmark in 2002 (source ANIMO database).

Country of origin	No. shipments	No. of portions
Netherlands	2	1,939
Sweden	2	40
Germany	86	1,982
Total	90	3,921

Wildlife: The role of wildlife in disease introduction is often difficult to assess or prove. However, Cape buffalo in southern Africa are believed to be a reservoir for FMDV, in particular the SAT types, and outbreaks in cattle are often blamed on incursions of buffalo into livestock areas (Thomson, 1994). Similarly, and of much more importance in Europe, wild boar (*Sus scofa scofa*) are a reservoir for CSFV. In Germany, an estimated 55% of primary CSF outbreaks in domestic swine herds have been attributed to contacts with wild boar (Fritzemeier et al., 2000).

Denmark currently does not have a free-ranging wild boar population although a small number are kept in fenced areas. The region of Germany north of the Kieler Canal that borders Denmark in Jutland has had a campaign to encourage wild boar hunting to keep the population down. Furthermore, any such animals crossing the border into Denmark are usually shot. In 2002, two wild boars were observed, one was shot at the border while the other managed to move 10km into Denmark and enter a swine herd and mate with some of the sows. A role for birds and rodents has been proposed for the transmission of both FMDV and CSFV but there is very little evidence to support this (Elbers et al., 2001). However, for reasons of general disease control and hygiene, rodents and birds should be excluded as much as possible from contact with farm animals and particularly with their feed, and this will clearly help reduce any potential risk of FMD and CSF.

Illegal Meat Imports: There has been considerable concern about the illegal importation of meat and animal products, particularly after the 2001 UK outbreak of FMD. This outbreak was thought to have been due to the illegal feeding of uncooked swill, which contained infected illegally imported meat. Illegal imports are by their nature very difficult to quantify, but there have been a number of attempts. Estimates from the UK suggest that there were some 7,431 (90% CI: 2,771-17,848) tonnes of illegally imported meat entering the UK in 2002 of which around 95kg was likely to be infected with FMDV. It is estimated that around 85.2% of this entered in personal baggage (DEFRA, 2003).

Table 8. Summary statistics from the routine surveillance of illegal meat imports at Kastrup airport, Denmark, between 23/01/03 and 26/06/03. This includes items intercepted in personal luggage and postal items that were seized.

Product description	Region	Total wt (kg)	No. of items	Avg. wt (kg)	Max. wt (kg)
Meat	Baltic Countries	3	1	3	3
Meat	Caribbean	41.5	6	6.9	30.2
Meat	Central America	2.9	1	2.9	2.9
Meat	Former Soviet Union	1.5	3	0.5	0.7
Meat	East Africa	97.3	13	7.4	30.1
Meat	Eastern Asia	190.9	49	3.8	66
Meat	Eastern Europe	126.6	43	2.9	9
Meat	Europe	6.0	1	6.0	6.0
Meat	Middle East	62.1	14	4.4	16.1
Meat	North Africa	22.3	4	5.5	8.9
Meat	South America	1.1	1	1.1	1.1
Meat	South-East Asia	199.6	33	6.0	47
Meat	West Africa	5.0	2	2.5	3.6
Meat/fish	Eastern Asia	4.2	1	4.2	4.2
Meat/egg	Eastern Asia	0.4	1	0.4	0.4
Meat/egg	Eastern Europe	1	1	1	1
Meat/Milk	Former Soviet Union	2.5	1	2.5	2.6
Meat/Milk	Middle East	7.5	1	7.5	7.5
Meat/Milk	North Africa	4.8	1	4.8	4.8
Meat/cheese	Caribbean	19.5	3	6.5	10.5
Meat/cheese	Former Soviet Union	11.2	3	3.7	4.4
Meat/cheese	East Africa	15.5	1	15.5	15.5
Meat/cheese	Eastern Asia	1.2	1	1.2	1.2
Meat/cheese	Eastern Europe	122.4	20	6.1	15.5
Meat/cheese	Europe	1.2	1	1.2	1.3
Meat/cheese	Middle East	13.3	2	6.6	8.4
Meat/butter	East Africa	7	1	7	7

As part of this assessment, the Customs at Copenhagen free port and Kastrup airport were contacted. The Danish port Customs use a computerised system for monitoring all imports. A random check is made on around 1% of containers passing through the port to confirm that they are correctly documented and to check for smuggling. The software also has some risk assessment components, and countries with known smuggling problems can be highlighted. From these records there was no evidence of illegal imports through the port. This possibly reflects the small size and ethnic mix of the immigrant communities in Denmark.

The Customs at the airport x-ray around 75% of the mail items entering the country and do spot checks on flights from countries where they have had problems of illegal imports. It was not possible to establish what proportion of passengers have their luggage searched. Based on seizures from luggage and from the mail over the first five months of 2003, it was estimated that 2,332kg of illegal animal products were detected entering Denmark annually, although this probably represents only a fraction of the total. These come mainly from China and Thailand in the Far East, and Ethiopia in Africa. However, this also reflects bias from previous experience since Customs target high risk flights. In future, it would be useful to improve the classification of seizures, and include an estimate of the numbers of items actually searched as well as the total numbers of items and people entering the country.

A small study was carried out in the airport in the summer of 2003, and all passengers from all flights arriving in Copenhagen from Thailand, Turkey, Syria, Ethiopia, Somalia, and Pakistan were searched. The summary of illegal animal products is given in Table 9. Over this two-week period, a total of 128.4kg of animal products were seized. If this is a representative sample, over the year there could be around 3,338kg of animal products entering Denmark illegally. As these countries are the main origins of illegal imports, this probably represents the majority of the imports in baggage. In this case the volume of illegal imports into Denmark was very small compared with the UK. However, one problem with this estimate is the exclusion of people from third countries who may have passed through another EU state before arriving in Denmark. These passengers do not appear in the sample and are not searched on entering Denmark.

The weights of individual items seized are generally small, with the majority of items weighing less than 8kg with one-third being less than 3kg. Therefore most of this material is likely to be for personal consumption. The risk that this poses depends on the species that the animal product came from, the type of processing undergone, if any, and the amount of time since the animal was slaughtered or the product was collected. Once in Denmark, products move along the exposure pathway, which is potentially very complex. In order to present a hazard, they must be infected, be thrown out and fed uncooked to pigs. This is extremely unlikely in Denmark since all swill feeding is now banned, most meat is likely to be cooked before any scraps are thrown out, and those scraps mostly end up in an incinerator. In the UK, it was estimated that only 0.013% of infected meat entering the UK was ingested by susceptible species, amounting to 12.5g. If the same assumption is made but using the estimate of 2,098kg of illegal meat (not including other products such as cheese) seized in a year, the amount of infected meat imported and fed to Danish livestock would be about 3.5mg. If it is assumed the oral ID_{50} is 10^5 $TCID_{50}$ (Sellers, 1971) and a gram of 'meat' has potentially 10.72 $TCID_{50}$ (Gale, 2002), then approximately 0.04 $TCID_{50}$ would be imported annually which represents a negligible risk. On the other hand, the oral ID_{50} is much smaller for CSFV, with as little as 10 $TCID_{50}$ needed to infect a pig (Farez & Morley, 1997). The viral loading in pork is estimated to be around $10^{4.9}$ $TCID_{50}$ per gram (Farez & Morley, 1997), therefore, a gram of pork could contain almost 8,000 ID_{50}. However, much of this meat would not be pork (as it comes from Islamic states) and the amount of infected meat reaching pigs would be much smaller, and therefore the risk of CSFV introduction via illegal meat imports into Denmark is low to negligible.

The swine herds most likely to be exposed to infected meat are those where the public can have contact with the pigs, for example those with outdoor pens, so that infected meat could be fed, either accidentally or intentionally. Efforts should be made to identify these holdings and ensure the operators are well informed of the hazards. In addition, where possible, they should be sited well away from other swine premises.

Table 9. Summary table of seizures made at Kastrup airport between 16/06/03 and 29/06/03 during an intensive campaign directed at illegal imports from Thailand, Turkey, Syria, Ethiopia, Somalia and Pakistan.

Product	Country	Region	Total wt. (kg)	No. items	Avg wt. (kg)
Butter	Ethiopia	East Africa	3.5	1	3.5
Cheese	Bosnia and Herzegovina	Eastern Europe	7	1	7
Cheese	Egypt	North Africa	3.6	1	3.6
Cheese	Jordan	Middle East	7.7	1	7.7
Cheese	Serbia and Montenegro	Eastern Europe	6	2	4
Cheese	Turkey	Middle East	19.9	5	7.7
Meat	Bosnia and Herzegovina	Eastern Europe	10.3	1	10.4
Meat	Ethiopia	East Africa	21.9	1	21.9
Meat	Indonesia	South-East Asia	0.6	1	0.6
Meat	Iran	Middle East	1.0	1	1.0
Meat	Lebanon	Middle East	6	1	6
Meat	Poland	Eastern Europe	3.0	1	3.1
Meat	Somalia	East Africa	2.0	1	2.1
Meat	Thailand	South-East Asia	9	2	6.5
Meat & Cheese	Ethiopia	East Africa	19.5	2	12.5
Sausage	Hungary	Eastern Europe	1.8	1	1.9
Sausage	Turkey	Middle East	5.6	1	5.6

Movement of People: The importation of FMDV and CSFV is also possible in fomites on people. This pathway involves the movement of people between farms within Denmark and other parts of Europe. Across Europe, farming is becoming increasingly intensive and in some cases, production has become dispersed across several countries for a single operation. In Denmark, several farmers are buying farms in the former Eastern Block countries that are about to join the EU, and they intend to have breeding herds in Denmark and fattening herds in other countries where labour is cheaper. There is also an increasing number of farm workers moving between units across national borders. This is a hazard for carrying disease into Denmark. Unfortunately, there is currently no information available on how frequently this practice is utilised. Hunters travelling to and from other EU states, particular Germany, to hunt wild boar present a similar type of problem. Further work is needed to understand better the potential hazards and also the scale of the problem.

CONCLUSIONS

This study is in no way exhaustive, but covers the major recognised routes of disease introduction of FMDV and CSFV and it has highlighted several other potential routes where more information should be sought. In general, the likelihood of introduction of FMDV is low with the main hazard coming from airborne spread. Of course, this needs to be reviewed as the patterns of trade and movements of people change. For CSFV on the other hand, there are several pathways of concern. This is mostly due the close proximity of CSF in Germany, the large number of trucks returning to Denmark and the import of boar semen. There is a low to negligible likelihood of introduction from illegal meat imports.

Currently, individual states within the EU are engaged in their own risk assessment activities, which is understandable. However, much of the information needed, particularly for quantitative assessments, is common to all EU states and a combined approach is needed as all the states share a common trade border.

ACKNOWLEDGEMENTS

This research was funded as a project of the International EpiLab (93S-2465-Å02-01437) Mark Bronsvoort conducted the research as a guest scientist of the International EpiLab in Denmark.

REFERENCES

Bachrach, H.L., Breese, S.S., Callis, J.J., Hess, W.R. and Patty, R.E. (1957). Inactivation of foot-and-mouth disease virus by pH and temperature changes and by formaldehyde. P.S.E.B.M. 95, 147-152

DEFRA (2003). Quantitative Risk Assessment for the Introduction of Foot-and-Mouth Disease in the United Kingdom.

de Vos, C.J., Horst, H.S. and Dijkhuizen, A.A. (2000). Risk of animal movements for the introduction of contagious animal diseases into densely populated livestock areas of the European Union. Proceedings of the Society for Veterinary Epidemiology and Preventive Medicine, Edinburgh, 124-136

Donaldson, A.I., Alexandersen, S., Sørensen, J.H. and Mikkelsen, T. (2001). Relative risks of the uncontrollable (airborne) spread of FMD by different species. Vet. Rec. 148, 602-604

Edwards, S. (2000). Survival and inactivation of classical swine fever virus. Vet. Microbiol. 73(2-3), 175-181

Elbers, A.R.W., Stegeman, A., Moser, H., Ekker, H.M., Smak, J.A. and Pluimers, F.H. (1999). The classical swine fever epidemic 1997-1998 in The Netherlands: descriptive epidemiology. Prev. Vet. Med. 42(3-4), 157-184

Elbers, A.R.W., Stegeman, J.A. and de Jong, M.C.M. (2001). Factors associated with the introduction of classical swine fever virus into pig herds in the central area of the 1997/98 epidemic in The Netherlands. Vet. Rec. 149(13), 377-382

Farez, S. and Morley, R.S. (1997). Potential animal health hazards of pork and pork products. Rev. Sci. Tech. Off. Inter. Epiz. 16(1), 65-78

Fritzemeier, J., Teuffert, J., Greiser-Wilke, I., Staubach, C., Schluter, H. and Moennig, V. (2000). Epidemiology of classical swine fever in Germany in the 1990s. Vet. Microbiol. 77(1-2), 29-41

Gale, P. (2002). Risk assessment: Use of composting and biogas treatment to dispose of catering waste containing meat. DEFRA. 182p

Gallagher, E., Ryan, J., Kelly, L., Leforban, Y. and Wooldridge, M. (2002). Estimating the risk of importation of foot-and-mouth disease into Europe. Vet. Rec. 150(25), 769-772

Greiner, M. (2003). The International Research Centre for Veterinary Epidemiology (International EpiLab) in Denmark. 10th Proceedings of the International Symposium for Veteirnary Epidemiology and Economics, Vina del Mer, Chile.

Hennecken, M., Stegeman, J.A., Elbers, A.R.W., van Nes, A., Smak, J.A. and Verheijden, J.H.M. (2000). Transmission of classical swine fever virus by artificial insemination during the 1997-1998 epidemic in The Netherlands: A descriptive epidemiological study. Vet. Q. 22(4), 228-233

Horst, H.S., Huirne, R.B.M. and Dijkhuizen, A.A. (1996). Eliciting the relative importance of risk factors concerning contagious animal diseases using conjoint analysis: A preliminary survey report. Prev. Vet. Med. 27(3-4), 183-195

Horst, H.S., Dijkhuizen, A.A., Huirne, R.B.M. and De Leeuw, P.W. (1998). Introduction of contagious animal diseases into The Netherlands: elicitation of expert opinions. Livestock Prod. Sci. 53(3), 253-264

Rugbjerg, H., Cheiel, M., Thomsen, L.K. and Willeberg, P. (1998). Risk assessment of importing classical swine fever to Denmark. Proceedings of the Society for Veteirnary Epidemiology and Preventive Medicine, 208-214.

Sellers, R. (1971). Quantitative aspects of the spread of foot and mouth disease. Vet. Bull. 41, 431-439

Sørensen, J.H., Jensen, C.O., Mikkelsen, T., Mackay, D.K.J. and Donaldson, A.I. (2001). Modelling the atmospheric dispersion of foot-and-mouth disease virus for emergency preparedness. Physics and Chemistry of the Earth Part B-Hydrology Oceans and Atmosphere 26(2), 93-97

Sutmoller, P. and Wrathall, A.E. (1997). A quantitative assessment of the risk of transmission of foot- and-mouth disease, bluetongue and vesicular stomatitis by embryo transfer in cattle. Prev. Vet. Med. 32(1-2), 111-132

Thomson, G.R. (1994). Foot-and-mouth disease. In: Infectious Diseases of Livestock with Special Reference to Southern Africa. J. A. W. Coetzer, G. R. Thomson and R. C. Tustin (Ed.). Oxford University Press, Oxford. 2, 823-852

Westergaard, J.M. (1982). Report on the eradication of foot-and-mouth disease on the islands of Funen and Zealand, Denmark 1982. Danish Veterinary Institute, Copenhagen. 97p

EVALUATION OF ZOONOSES CONTROL PROGRAMMES FOR ANIMAL AND PUBLIC HEALTH WITH A DYNAMIC MODEL: THE EXAMPLE OF LIVESTOCK BRUCELLOSIS VACCINATION IN MONGOLIA

J. ZINSSTAG*, F. ROTH, D. ORKHON, G. CHIMED-OCHIR, M. NANSALMAA AND P. VOUNATSOU

SUMMARY

We report here a dynamic model of livestock to human brucellosis transmission which was developed as the underlying framework to estimate the profitability and cost-effectiveness of brucellosis control in Mongolia. The compartmental model considers transmission within sheep and cattle populations and the transmission to humans as additive components. The model was fitted to demographic and sero-prevalence data (Rose Bengal Test) from livestock and annually newly reported human brucellosis cases in Mongolia for a period of ten years (1991-1999) prior to the onset of a mass livestock vaccination campaign (S19 *Brucella abortus* for cattle and Rev1 *B. melitensis* for sheep and goat). The vaccination effect was fitted to livestock and human disease data from the first three years of the mass vaccination campaign (2000 -2002). Parameters were optimized on the basis of the goodness of fit, which was assessed by the deviance. Average effective reproductive ratios for the year 1999 are 1.15 for sheep and 1.67 for cattle indicating a very low, slow growing transmission. In general the model mechanism is able to fit and simulates plausibly the different vaccination and test and slaughter interventions and their respective effect on public health. The economic analysis of the vaccination campaign (parallel paper) indicates a Benefit Cost Ratio for society of 3.2 (min. 2.27; max. 4.37).

INTRODUCTION

Brucellosis is one of the world's major zoonoses (Boschiroli et al., 2001) Its importance is often not well known, but it can have a considerable impact on human and animal health, and wide socio-economic impacts especially in countries where rural income relies to a large extent on livestock breeding and dairy products. Human brucellosis is commonly caused by exposure to infected livestock and livestock products (mostly raw milk and milk products). The most important causative bacteria in decreasing order are: *Brucella melitensis* (small ruminants), *B. abortus* (cattle), *B. suis* (pigs) and *B. canis* (dogs). There is no transmission of the infection between humans (Krauss et al., 1996) but humans may infect animals (Parnas et al., 1966). In humans, mortality is reported to be negligible, but the illness can last for several years. (Madkour, 2001) In animals, brucellosis mainly affects reproduction and fertility, reduces

---

* Jakob Zinsstag, Swiss Tropical Institute, PO Box, 4002 Basle, Switzerland
Email: jakob.zinsstag@unibas.ch

survival of newborns and reduces milk yield. Mortality of adult animals is insignificant (Sewell & Brocklesby, 1990).

Control strategies available to prevent human infection are pasteurisation of milk, livestock vaccination and the elimination of infected animals. In Mongolia, in the 1970s mass vaccination of livestock successfully reduced human annual incidence to less than one case per 10 000 (Kolar J., pers. commun.). After democratic reform, and the shift away from dependence on the former Soviet Union in 1990, human brucellosis re-emerged as a major, but preventable, source of illness. Based on recommendations made to the World Health Organisation (WHO) and the Mongolian government (Garin-Bastuji, 1999), a whole-herd vaccination strategy covering 10 years was developed (Mikolon, 2000) to start in 2000.

Very little is known about the economic implications of brucellosis and brucellosis control for human health in any country (Bernues et al., 1997). The zoonotic nature of brucellosis needs a multi-sectorial assessment, including human health, the socio-economic situation of the concerned population groups and livestock production. Edmunds et al. (1999) recommend a dynamic assessment of the economic effects of vaccination campaigns, as static approaches underestimate its cost-effectiveness because of the changing force of infection. Available models of brucellosis transmission consider only different livestock species although brucellosis is transmitted to humans from small ruminants (sheep and goat) and cattle (Roe, 1977; Carpenter, 1987; Dalrymple, 1993; Gonzalez-Guzman et al., 1994). We report here a dynamic model of livestock to human brucellosis transmission underlying the cost effectiveness analysis. The economic part of the cost-effectiveness to human health and potential net economic benefits of a nation-wide mass vaccination programme for livestock over a period of 10 years is reported elsewhere (Roth et al., 2003).

MATERIALS AND METHODS

Data collection

Livestock demographic data where provided by the Mongolian Statistical Office. (Mongolian Statistical Yearbook, 1999). Animal brucellosis sero-prevalence survey data (Rose Bengal) were provided on the provincial level for cattle and sheep for the years 1990 – 1999 by the Ministry of Agriculture for the model fit without intervention. The sero-prevalence in cattle and sheep varied between 0.5 and 3% The standard errors of the sero-prevalence estimates did not exceed 0.045% in cattle and 0.035% in sheep. Data on the ongoing mass vaccination campaign was provided from 2000 – 2002. The quality of the available demographic and sero-prevalence data presented could not be checked, but ongoing studies on brucellosis in livestock indicate that the reported sero-prevalence is underestimated (A. Mikolon, pers. comm.) Our analysis is thus rather conservative and was restricted to apparent sero-prevalences provided by the Rose Bengal test. Baseline disease data on reported cumulative incidence of human brucellosis listed by Province (Mongolian: Aimag), for the years 1990-1999 was provided by the Infectious Disease Research Institute (IDRI) in Ulaanbaatar. The human brucellosis data are annually newly reported human cases who where diagnosed clinically and confirmed by various serological tests (Rose Bengal, Huddelson, Wright). These tests do not seem well standardised between the different Mongolian provinces. However, Mongolian physicians have longstanding and extended experience with brucellosis. Brucella cultivation from human patients is rarely done.

Model

We developed a deterministic model of animal to human brucellosis transmission in steps of one year which is adapted to the availability of data for validation (Roth et al., 2003). Because only data on seropositive animals was available, we used, instead of two "infectious" and "recovered" compartments, only one "seropositive" compartment. We consider transmission between sheep, between cattle, and from both livestock species to humans. We omitted the transmission from goats due to the lack of data. Sheep total population data is only available for the years 1995 to 1999. We reconstructed the total population for the years 1990 to 1994 by linear regression on the years 1995 to 1999. Brucellosis affects mostly fertility and milk production. In the model we consider a sero-prevalence dependent effect on fertility for sheep and cattle in (Bernues et al., 1997). The incidence is calculated by the product of the proportion of infectious, expressed as uniform probability distribution, the contact rate, the number of susceptible the number of sero-positive animals.

For the human susceptibles, we considered the whole Mongolian population, as precise estimates of the population at risk are not available. A second compartment contains the annually newly registered brucellosis patients and third compartment represents the registered patients between year 2 and 3 of state registration. The model here takes Mongolian health policy into account. This is important to adapt the assessment to the Mongolian reality of health financing. We refer to Habicht et al. (1999) who recommend to adapt health systems analysis to the expectations and decision pathways of health authorities in particular countries. With a view to extending the model, e.g., to adapt the model to other contexts, the duration of untreated human brucellosis was obtained from the analysis of data by Beklemischew (1966) on a thousand human brucellosis patients in Kazakhstan. The distribution of the duration of clinical brucellosis fits best with an exponential decay function with an average number of 3.11 disease years (Minimum <1 year, Maximum 18 years) R-square = 0.996 (Zinsstag et al., 2004). Considering the reported persistence of antibodies until 10 years post end of clinical cure reported by Wundt et al. (1968), the loss of human post infection immunity, could be set at 0.1.

Fitting the transmission model

The fitting of the model to the data was done with Vensim(TM) systems analysis software (Ventana Systems Inc., 60 Jacob Gates Road Harvard MA 01451, USA; www.vensim.com) using the Powell nonlinear maximum likelihood optimization algorithm (Press et al., 1991). Parameters where optimized on the basis of the goodness of fit, which was assessed by the deviance. In a first step, mortality and birth rates were optimized for the susceptible sheep, cattle and humans. Birth rates were expressed proportionally to the total populations and mortality parameters in livestock include natural mortality and off take. In a second step the transmission within sheep and within cattle were fitted by fixing the demographic parameters. To fit the transmission process, the proportions of infected sheep and cattle were expressed as uniform probability distributions and their boundaries were varied to identify the best fit in terms of the deviance of contact rates for the transmission between sheep and between cattle and the loss of immunity. The transmission to humans is expressed as additive contributions of transmission from sheep and cattle to humans. The effective reproductive ratio and threshold vaccination coverage were estimated according to Scott and Smith (1994).

Simulation of vaccination and test and slaughter scenarios

The current practice in Mongolia (since 1990) consists of low-level surveillance, with occasional testing of livestock herds followed by voluntary slaughter sero-positive animals without compensation. The present vaccination strategy aims at vaccinating all adult animals twice within 6 years (1/3 of the total adult population per year). All animals born during the 10 years of the plan will be vaccinated once (at < 1 year of age). The reported efficacy in reducing transmission is considered as the prevented fraction (1 – R), where R is the relative risk of disease in those who receive the intervention compared to those who do not (Smith & Morrow, 1991). The vaccines to be used in cattle (Strain B19, *Brucella abortus*) and small ruminants (Rev 1, *Brucella melitensis*) should reduce transmission by at least 65% (Nicoletti, 1977). These efficacies also include potential losses due to cold chain deficiency (Mikolon, pers. comm.). Vaccine coverage scenarios were set at 50% and 80%. The proportion of protection PP is computed as the product of the reduction of transmission RT of the respective vaccine and the vaccination coverage VC as vaccinated young and adult sheep (Rev1 *B. melitensis* sheep vaccine) and young and adult cattle (S19 *B. abortus* cattle vaccine) are the product of vaccine efficacy and vaccination coverage.

Three different vaccination scenarios where considered assuming PP of 32% (RT= 65%; VC= 50%); 52% (RT= 65%; VC= 80%) and 80% (RT= 100%; VC= 80%), respectively. For Rev 1 we considered an annual loss of vaccination immunity as a random function (Uniform (0.2, 0.25, inverse of duration of vaccination immunity of 4-5 years, WHO 1998) and for S19 an annual loss of vaccination immunity as a random function (Uniform (0.125, 0.142; inverse of duration of vaccination immunity of 7-8 years, (AFSSA, 2001)). FAO suggested including the testing and slaughtering of sero-positive animals as a separate intervention. For the simulation, the current capacity of livestock Rose Bengal testing of the Mongolian veterinary laboratories was doubled to consider 40% of the national livestock population being tested and the sero-positive animals removed.

RESULTS

Model fit

The model fitted the susceptible livestock and human population very well. For the fit of the transmission between sheep a proportion of infectious sheep among the seropositive sheep ranging between 0.2 and 0.8 (uniform) yielded the best fit. In cattle the best fit was found for a proportion of infectious animals among the seropositives ranging between 0.1 and 0.85. The simultaneously fitted sheep-human and cattle-human contact rate shows a contribution of sheep derived human brucellosis of over 90% indicating a dominance of *B. melitensis* in human brucellosis. The contact rate for cattle to human transmission is 2.11e-9 (95% Confidence Interval: 2.09e-9; 2.13e-9) and for sheep to human transmission is 1.127e-8 (95% Confidence Interval: 1.124e-8; 1.128e-8). Reproductive ratios for within sheep and within cattle transmission were calculated using the confidence limits of the proportions of infectious animals obtained during the fitting process. Average effective reproductive ratios for the year 1999 are 1.15 for sheep and 1.67 for cattle indicating a very low, slow growing transmission. Maximum threshold vaccination coverages are 0.46 for sheep and 0.66 for cattle.

Simulation of interventions

Vaccination: For the economic analysis of brucellosis mass vaccination in Mongolia we simulated the different vaccination scenarios for ten years and use the differences of the disease data with and without interventions as inputs for the health and livestock economic assessment (Roth et al., 2003). A sensitivity analyses was done by assigning probability distributions to the model parameters based on the confidence limits of the fit (Roth et al., 2003) In sheep the reported sero-prevalences for the years 2000 – 2002 tally best between the scenarios of 52% and 80% protection. In cattle the simulated vaccination scenarios follow the reported trend for the years 2000 to 2002 but at a higher level.

Test and Slaughter: The simulation of the Rose Bengal testing of 40% of the sheep and the removal of the seropositives results in a comparable decrease as in the vaccination scenarios. The test and slaughter intervention in cattle appears more effective to reduce brucellosis prevalence in cattle than the vaccination scenarios. However, the simulated vaccination and test and slaughter intervention scenarios lead not to eradication after 10 years.

DISCUSSION

Model

To the best of our knowledge, the reported model is the first comprehensive dynamic assessment of livestock to human brucellosis transmission fitted to a period of transmission with and without intervention. The model conception was adapted to the available data (only sero-prevalence data), the needs of economic assessment (Roth et al., 2003) and the consideration of the Mongolian brucellosis patient registry policy to adapt the analysis to the needs of the Mongolian authorities (Habicht et al., 1999). The fit to the data is satisfactory but the model captures only the large trends of the diseased compartments of livestock and humans and not the smaller modulations. The proportions of infectious sheep and cattle may in fact range in more narrow bands (Bürki and Sackmann, pers. Comm.. estimated it between 50-60%), but the presented range yielded the best fit and is in line with Parnas (1966) who estimates the range of excreters between 22-60%.

Interventions

In general, the model mechanism is able to fit and simulate the different interventions plausibly. However, the long time step causes oscillations and a lag period of the effect on human disease. The model predicts a decrease of sero-prevalence in livestock by the vaccination campaign. After ten years, the intervention strategies should be reviewed to consider that brucellosis may remain only in certain areas. In cattle, a test and slaughter policy should be considered at the end of the mass vaccination campaign, if there is compensation for slaughtered animals. The model can produce estimates of the numbers of animals for which compensation is required. The vaccination campaign seems more effective in reducing prevalence in sheep than in cattle. For the economic analysis we use the scenario of 52% protection, which resulted in a benefit-cost ratio of 3.2 (min. 2.27; max. 4.37) for the whole society (Roth et al., 2003).

Limitations of the model

We did not consider neither geographical differences of disease occurrence, age dependence, nor could we include goats which may play an important role in the transmission to humans. The model could become smoother by reducing time steps to half year or quarter intervals. This would probably also reduce the overestimation of the vaccinated animals in our model. We should bear in mind that the quality of the demographic and sero-prevalence data is unknown.

Further research

More research is needed to estimate the proportion and duration of brucella excreting small ruminants and cattle among sero-positives. An adaptation of the model concept to rabies transmission is ongoing. The model could be used to assess other zoonoses and food borne diseases. Improvements of the model should include spatial effects and may be formulated using Bayesian Markov Chain Monte Carlo methodology (Marshall et al., 2003)

ACKNOWLEDGEMENTS

We would like to thank Ms. Oyunaa Tsendendorji and collaborators at the Ministry of Health, in Ulaan Baatar, for their help throughout our stay in Mongolia, for arranging the timetable and logistical support, for organising the studies, for entering the data and serving as a competent interpreter and friendly guide. We would also like to thank all the collaborators of the Ministry of Agriculture and Industry and of the Ministry of Health for their hospitality and their sound introduction to the complex topic of brucellosis in Mongolia. Data was provided by D. Bat-Ochir and D. Idesh, National Centre of Infectious Diseases (Ministry of Health), D. Nyamkhorol, Statistical Information Department, Directorate of Medical Services; P. Dorjsuren and P. Bolortuya, State Veterinary And Animal Breeding Department; A. Yondondorj, Veterinary Research Institute, all in Ulaan Baatar, Mongolia. Tumurkhuu Gantsetseg provided information on clinical aspects of brucellosis in Mongolia. Many thanks to the staff of WHO/Mongolia for their hospitality and especially to R. Salmela for assisting us by introducing us to the health system in Mongolia. M. Dubach is acknowledged for providing background information. D. Pfeiffer contributed to the initial design of the study. A. Mikolon gave substantial technical inputs to brucellosis control strategies and intervention budgets. J. Kolar contributed to the conception of the study from his experience in running mass vaccination campaigns against brucellosis. J. Nicolet, W. Sackmann, P. Nicoletti and F. Bürki provided information on the proportion of infectious animals. Funding: World Health Organisation, Food and Agriculture Organisation, Swiss Tropical Institute. JZ was funded by the National Center for Competence in Research North South, Individual Project 4 (NCCR North-South IP4).

REFERENCES

AFSSA, Maisons Alfort (2001). La brucellose animale. http://www.afssa.fr

Beklemischew, N.D. (1966). Heilung. Reinfektion. Verlauf der Brucellose beim sensibilisierten Organismus. In: Die Brucellose des Menschen (Human Brucellosis) Parnas J. Krüger W. Töppich E. eds. VEB VErlag Volk und Gesundheit, Berlin 566pp

Bernues, A., Manrique, E. and Maza, M.T. (1997). Economic evaluation of bovine brucellosis and tuberculosis eradi-cation programmes in a mountain area of Spain. Prev. Vet. Med. 30,137-149

Boschiroli, M.L., Foulogne, V. and O'Callaghan, D. (2001). Brucellosis: a worldwide zoonosis. Current Opinion in Microbiol. 4, 58-64

Carpenter, T.E., Berry, S.L. and Glenn, J.S. (1987). Economics of Brucella ovis control in sheep: epidemiologic simulation model. J. Am. Vet. Med. Assoc. 190, 977-982

Dalrymple, M. (1993). Model for assessing the risk of introducing brucellosis into a brucellosis-free area. Rev. Sci. Tech. 12, 1175-1186

Edmunds, W.J., Medley, G.F. and Nokes, D.J. (1999) Evaluating the cost-effectiveness of vaccination programmes: A dynamic perspective. Stats in Med. 18, 3263-3282

Garin-Bastuji, B. (1999). Report of a mission to Mongolia from 2 to 16 April 1999, on the "Epidemiological surveillance and control of communicable diseases of public health importance, including zoonoses".World Health Organization, Geneva, p5.

Gonzalez-Guzman, J. and Naulin R. (1994). Analysis of a model of bovine brucellosis using singular perturbations. J. Math. Bio. 33, 211-223

Habicht, J.P., Victora, C.G. and Vaughan. J.P. (1999). Evaluation designs for adequacy, plausibility and probability of public health programme performance and impact. Int. J. Epidemiol. 28,10-8

Krauss, H., Weber, A., Enders, B., Schiefer, H.G., Slenczka, W. and Zahner, H. (1996). Zoonotic diseases, infection diseases transmitted from animals to human (Original title in German: Zoonosen, von Tier auf den Menschen übertragbare Infek-tionskrankheiten. 2nd. Edition. Deutscher Ärzte-Verlag Köln 400pp

Madkour, M.M. (1984) Madkour's Brucellosis. Springer Verlag, Berlin Heidelberg 2001:306 pp

Marshall, E.C., Frigessi, A., Stenseth, N.C., Holden, M., Ageyev, V.S. and Klassovskiy, N.L. (2003). Plague in Kazakhstan: a Bayesian model for the temporal dynamics of a vector-transmitted infectious disease. Technical Report, Dept. of Epidemiology and Public Health, Imperial College, London. http://www.med.ic.ac.uk/divisions/60/biostat/plague.ps

Mikolon, A. (1999). Report of the WHO mission to Mongolia from 14. August to 3. No-vember World Health Organization, Geneva. p74

Mongolian Statistical Yearbook (1999). Mongolian Statistical Office, Ulaan Baatar, Mongolia

Nicoletti, P. (1977), Adult vaccination. In: Crawford RP and Hidalgo RJ (eds.) Bovine Brucellosis. Texas A&M University Press, college Station, TX 1977, 177-188

Parnas, J. Krüger, W. And Töppich, E. (1966). Die Brucellose des Menschen (Human Brucellosis) VEB Verlag Volk und Gesundheit, Berlin 566pp

Press, W.H., Flannery, B.P., Teukolsky, S.A. and Viatour, P. (1991). Numerical Recipes in C. Cambridge: p309

Roe, R.T. (1977), The application of computer simulation to the planning of the public investment in the control of animal disease. PhD Thesis, Faculty of Veterinary Sciences, University of Melbourne, Australia

Roth, F., Zinsstag, J., Orkhon, D., Chimed-Ochir, G., Nansalmaa, M., Hutton, G., Cosivi, O., Carrin, G. and Otte, J., (2003). Human health benefits from livestock vaccination for brucellosis: a case study. Bulletin of the World Health Organization 81, 867-876

Scott M.E. and Smith G. (1994). Parasitic and infectious diseases : epidemiology and ecology. New York (USA): Academic Press

Sewell, M.M.H. and Brocklesby, D.W. (1990). Animal diseases in the tropics 4th Edition, Baillière Tindall, London 385pp

Smith, P.G. and Morrow, R.H. (1991). Methods for Field Trials of Inter-ventions Against Tropical Diseases, New York: Oxford University Press. 326pp

WHO (1998). The Development of New/Improved Brucellosis Vaccines. World Health Organization. WHO/EMC/ZDI/98.14

Wundt, W (1968). Infektionskrankheiten, Vol II(1), In Gsell, O. and Mohr, W. Springer Verlag, Berlin, Library of Congress Catalog Card Number 67-15610. p483-542

Zinsstag, J., Roth, F., Nansalmaa, M., Vounatsou, P., Schelling, E. and de Savigny, D. (2004). A contribution to the duration and disability weight of human brucellosis. Population Health Metrics (submitted)

AN EVALUATION OF THE DUTCH CLASSICAL SWINE FEVER SURVEILLANCE PROGRAMME

D. KLINKENBERG*, M. NIELEN, M.C.M. MOURITS AND M.C.M. DE JONG

SUMMARY

This paper presents an evaluation of the effectiveness of four Dutch classical swine fever (CSF) surveillance programmes: routine gross pathology, routine virological testing of tonsils, clinical observation by the farmer and periodic clinical inspections by veterinarians. The high-risk period (HRP), which was defined as the time between virus introduction and first positive diagnosis, was simulated with stochastic models of virus transmission, clinical disease development, and actions and decisions in surveillance programmes. As a measure for surveillance programme effectiveness, the number of infected herds at the end of the HRP was used. Also, the yearly costs of the programmes were calculated, and the consequential costs of an epidemic were related to the number of infected herds at the end of the HRP. The most important conclusions were that the current surveillance programmes can prevent expensive epidemics (> €100M) with high probability (75%) and that periodic clinical inspections may need to be reconsidered.

INTRODUCTION

Epidemics of classical swine fever (CSF) can result in major economic and social-ethical consequences, which are related to the number of infected herds at the end of the high-risk period (HRP). The HRP is the time interval in which the virus is spreading before awareness of the introduction of the classical swine fever virus (CSFV). To minimise the number of infected herds at the end of the HRP, surveillance programmes are generally utilised.

Continuation or improvement of surveillance programmes should depend on their effectiveness. Although the effectiveness of some Dutch surveillance programmes has been evaluated (Crauwels et al., 1999; Stegeman et al., 2000), a comparative study of multiple programmes has never been carried out. In this paper, an analysis of the effectiveness of four CSF surveillance programmes in The Netherlands is presented: gross pathological examination of pigs submitted to the Animal Health Service (AHS), virological testing of tonsil of pigs submitted to the AHS, daily clinical observation by the farmer and four-weekly clinical inspection by a veterinarian.

The effectiveness of a surveillance programme is a balance between the costs per year and the consequential costs during an epidemic if the programme is not used. Since all sorts of consequences (epidemiological, economic and social-ethical) depend on the number of infected

---

* Don Klinkenberg, Farm Animal Health, Utrecht University, Yalelaan 7, 3584CL Utrecht, The Netherlands. E-mail: d.klinkenberg@vet.uu.nl

herds at the end of the HRP, this number was determined as the best measure of programme effectiveness.

MATERIALS AND METHODS

The number of infected herds at the end of the HRP was determined with stochastic simulation models. Many combinations of surveillance programmes were considered as well as possible improvements. The models were also used to estimate the yearly costs of the programmes. Finally, simulated epidemics were used (Mangen, 2002) to examine the relation between the number of infected herds at the end of the HRP and the direct control costs of a CSF epidemic.

The models

Three model types were used to obtain the final results. Firstly, an epidemic model was used to simulate the transmission of CSFV within and between herds. With the epidemic model, 100 epidemics were simulated with complete information on the times of infection of all animals. Secondly, a disease model was used to simulate the development of clinical symptoms. With the disease model, the development of disease was simulated for all infected animals in all 100 epidemics. Thirdly, surveillance programme models were used to simulate the actions and diagnostic tests in the 100 epidemics with disease development. This resulted in a day of detection (the end of the HRP) and thus in a number of infected herds at the end of the HRP. The surveillance programme models were also used to calculate the yearly costs of the programmes.

The epidemic model

The epidemic model of Klinkenberg et al. (2003) was used with an adjustment of some of the parameter values since the original model described CSFV transmission where disease control measures were already in place. In the model, two herd types were present, piglet producing and selling herds (multiplier herds) and all other herds (finishing herds). The herd types differed with respect to contact rates, as piglets were only transported from multiplier herds to finishing herds, but did not differ with respect to within-herd transmission. Each herd was assumed to consist of 1000 pigs.

Virus transmission within herds was simulated with a stochastic SIR model, in which all animals were in one of the classes S (susceptible), I (infectious) or R (recovered). Each S animal could move to I with rate $\lambda i/n$, where i/n is the fraction of infectious animals in the herd, and each I animal could move to R with rate μ. The parameter values used were $\lambda = 0.21$ and $\mu = 0.074$ (Klinkenberg et al., 2003).

Virus transmission between herds was simulated with a branching process model, which implied that the number of susceptible herds was assumed to be unlimited. From infectious multiplier (*m*) and finishing (*f*) herds, new infectious herds ‘branched off’ with rates from the following transmission matrix:

$$\begin{array}{ccc} & m & f \\ m & \beta i(t) & \beta i(t) \\ f & (\beta+\gamma) i(t) & \beta i(t) \end{array}$$

in which $i(t)$ is the number of infectious animals in the infectious 'stem' herd, β is the between-herd transmission parameter for all contacts except piglet transportation, and γ is the transmission parameter for piglet transportation only. The used parameter values were β = 0.0024 and γ = 0.0029 (Klinkenberg et al., in prep.).

The disease model

Clinical disease was simulated at the level of the individual pig. Three disease levels were defined (aspecific, specific and severe disease) and, for each pig, random numbers were drawn so as to determine which disease level(s) it would be allocated. These probabilities and the period during which the animals were diseased were derived from 41 experimental infections with the CSFV strain of the Dutch 1997-1998 epidemic carried out at the Animal Science Group in Lelystad and are defined below:

1. Aspecific disease. Animals showed aspecific disease, consisting of symptoms such as fever, apathy, anorexia or diarrhoea with a probability of 88%. Consequently, 12% of the animals did not show any disease. Aspecific disease occurred from day 7 to 27 after infection of the animal.

2. Specific disease. Pigs with aspecific disease showed specific disease with a probability of 42%. Specific disease consisted of CSF specific symptoms such as conjunctivitis, skin haemorrhages, cyanotic ears or lameness, and occurred from day 12 to 27 after infection of the pig.

3. Severe disease. Pigs with specific disease showed severe disease with 80% probability, and animals with aspecific disease (but not specific disease) with 14% probability. Severe disease consisted of fever above 41ºC and at least 4 (specific or aspecific) symptoms for 3 days, or death. It occurred from day 13 to 27 after infection.

The surveillance programme models

The surveillance programme models consisted of chronological descriptions of actions, decisions and diagnostic tests. The models were applied separately to each of the 100 epidemics to determine the end of the HRP and thus the number of infected herds at the end of the HRP. To determine the result of simultaneous application of programmes, for each epidemic the earliest HRP end of the individual programmes was the end of the HRP for the compound programme.

The *pathology* programme: The *pathology* programme started when a veterinarian submitted a pig for gross pathological examination. The starting day was distributed with the following cumulative submission probability distribution: 20% probability on the first day with 5 severely diseased animals then, 30%, 40%, …, 80% on the subsequent six days with 100% on the eighth day.

Pathological examination was assumed to occur on the day of submission, and led to CSF suspicion with ±40% probability (Elbers et al., 2003), otherwise a new submission followed one week later. Suspicion led to quarantine measures and a tonsillar test on the same day, which was positive with 60% probability (Bouma et al., 2001) with a positive tonsillar test being the end of the HRP. If negative, new tonsillar samples (max. 5) and EDTA blood samples were taken one day later. The tonsillar samples were tested the same day with 60% sensitivity per tonsil and a positive tonsil indicating the end of the HRP. Negative tonsils resulted in a further four day delay until the virus isolation results from the EDTA blood samples (100% sensitivity) resulted in the end of the HRP (five days after initial submission).

The *tonsil* programme: The *tonsil* programme concerned the same animals as the *pathology* programme. Tonsils of all pigs were submitted for virological testing, which was carried out two days later. The tonsil was positive with 60% probability, resulting in the end of the HRP, if positive.

The *farmer* programme: The *farmer* programme started when the farmer observed (not recognised) CSF symptoms and called a veterinarian. The starting day was distributed with the following cumulative calling probability distribution: 20% probability on the first day with 5 specifically diseased animals, 30% on the first day with 6 specifically diseased, 40% with 7, …, and 100% on the first day with 13 specifically diseased animals.

The veterinarian visited the farm one day after the phone call and had a 32% probability of suspecting CSFV infection (Elbers et al., 2003). If the result was negative, he returned to the farm one week later. With suspicion, the farm was put into quarantine and tonsillar samples (max. 5) and EDTA blood samples were taken the same day, as in the *pathology* programme.

The *inspection* programme: The *inspection* programme started at a random day within the first 28 days after the introduction of infection onto a farm. A veterinarian visited the farm that day, and from there, the programme was equal to *farmer*. As an alternative programme, a two-weekly inspection was simulated.

Table 1. Estimated costs and frequencies of actions, decision, and diagnostic tests in the surveillance programmes in CSF-free years.

Action (price unit)	Estimated price per unit	Surveillance programme	Estimated frequency per year
Gross pathology (per pig submission)	€400	Pathology	4,500
Sampling on farms + sample analysis (per farm visit)	€3,000	*Pathology*	1
		Farmer	4
Tonsil virology (fixed price per year)	€166,000	*Tonsil*	1
Inspection (per farm per year)	€1,175	*Inspection_4wk*	13,000
	€2,085	*Inspection_2wk*	13,000
Quarantine (per farm visit)	€170	*Pathology*	1
		Farmer	4

Calculation of the yearly costs of the surveillance programmes

The costs of the surveillance programmes in CSF-free years were calculated from the costs and frequencies of all actions. Information was obtained from the websites of the AHS, the Central Institute for Animal Disease Control (CIDC), and the National Inspection Service for

Livestock and Meat (RVV). Table 1 shows all estimated costs and frequencies for the different actions and indicates the associated programme.

<u>The number of infected herds at the end of the HRP and the costs of a CSF epidemic</u>

Data from 200 stochastic simulations of epidemics were available (Mangen, 2002; Mangen et al., 2002). The simulations were used to examine the relationship between the number of infected herds at the end of the HRP and the direct costs of the epidemic i.e. the costs related to control of the epidemic and to compensating pig farmers. The simulated control scenario consisted of the culling of pigs on detected farms, contact tracing, establishment of protection and surveillance zones, and preventive culling of farms within 750 to 1000 metres around infected farms, which is the current standard Dutch control programme without vaccination.

RESULTS

Table 2 shows the results for all combinations of the four surveillance programmes.

Table 2. Simulated results for the four current Dutch surveillance programmes.

pathology	*tonsil*	*farmer*	*inspection*	# infected herds at end of HRP				
				5%	25%	**50%**	75%	95%
+				1	6	**13**	33	64
	+			1	4	**10**	17	42
		+		1	6	**14**	41	88
			+	4	17	**70**	139	251
+	+			1	4	**8**	13	34
+		+		1	4	**8**	13	43
+			+	1	4	**10**	23	60
	+	+		1	4	**8**	12	34
	+		+	1	4	**9**	16	42
		+	+	1	5	**11**	32	70
+	+	+		1	4	**6**	10	19
+	+		+	1	4	**7.5**	12	34
+		+	+	1	4	**7**	12	39
	+	+	+	1	4	**7**	12	26
+	+	+	+	1	4	**6**	10	19

It appears that application of all four programmes results in less than 19 infected herds at the end of the HRP with 95% probability, and in less than 10 infected herds with 75% probability. Of the individual programmes, *tonsil* is the most effective, followed by *pathology* and *farmer*. Applying those three programmes together is as effective as applying all four programmes together, which is in accordance with the smallest effectiveness of *inspection* only. Increasing the inspection frequency to once every two weeks does improve the effectiveness of *inspection* only, but not in combination with the other three programmes (results not shown).

The estimated yearly costs of the surveillance programmes are shown in Table 3. Since some costs are made irrespective of CSF surveillance (gross pathological examination in *pathology*, and some of the inspection visits by veterinarians in *inspection*), a separate column is added with only CSF specific costs. It appears that *pathology* and *farmer* are by far the cheapest programmes, *tonsil* is much more expensive, but not as expensive as *inspection*.

Finally, Table 4 shows the relation between the number of infected herds at the end of the HRP and the simulated direct costs of an epidemic. Costs will very likely remain below €100 million if the number of infected herds is below 10 and probably become larger than €150 million if the number of infected herds gets larger than 20.

Table 3. Estimated yearly costs of the surveillance programmes.

Programme	Costs per year (K€)	CSF specific (K€)
Pathology	1,800	3.17
Tonsil	166	166
Farmer	12.7	12.7
Inspection_4wk	15,300	12,500
Inspection_2wk	27,100	21,600

Table 4. Results of 200 simulated epidemics, indicating the relationship between the number of infected herds at the end of the HRP and the direct costs (from Mangen, 2002).

	Number of infected herds at the end of the HRP							
Direct costs	1-5 herds	6-10 herds	11-15 herds	16-20 herds	21-25 herds	26-35 herds	36+ herds	Total number of simulations
0-50 M€	17	6	2	0	0	0	0	25
50-75 M€	5	15	7	4	0	0	0	31
75-100 M€	0	11	15	11	8	2	0	47
100-125 M€	0	5	3	10	4	3	1	26
125-150 M€	0	1	2	8	3	8	0	22
150-200 M€	1	2	1	2	8	9	5	28
200+ M€	0	0	3	0	5	3	10	21
Total number of simulations	23	40	33	35	28	25	16	200

DISCUSSION

The effectiveness of four current CSF surveillance programmes in The Netherlands was evaluated. The effectiveness, measured as the number of infected herds at the end of the HRP, was compared to the yearly costs of the programmes. Finally, the relation between the number of infected herds at the end of the HRP and direct costs of an epidemic were examined.

It appeared that application of the two cheapest programmes, *pathology* and *farmer*, was more effective than the more expensive programmes, *tonsil* or *inspection*. However, if *tonsil* was added as a third programme, the 95-percentile was reduced from 19 to 9, which made *tonsil* seem a very useful surveillance measure. *Inspection* was the most expensive programme and did not improve the effectiveness of surveillance when added to *pathology*, *farmer* and *tonsil*. Moreover, even increasing the inspection frequency to once every two weeks was not effective.

The CSF simulations by Mangen (2002) revealed that less than 10 infected herds at the end of the HRP very likely leads to direct costs below €100 million (Table 4). When more than 20 herds are infected, epidemics will probably cost more than €150 million. The simulated CSF surveillance programmes kept the number of infected herds below 19 with 95% probability, and below 10 with 75% probability, so there is currently only a small probability of very expensive epidemics.

Leaving out the *pathology* programme would save €3,170 per year, and would increase the 50-percentile from 6 to 7 and the 95-percentile from 19 to 26. Consequently, the direct costs of an epidemic would roughly increase by 25 M€, so that leaving out *pathology* would be cost-effective if CSFV introductions occurred less than once in 8,000 years. Likewise, omitting *tonsil* would increase direct costs by ±50 M€, which is cost-effective if CSFV introductions were only once every 300 years. Omitting *farmer* increased direct costs by ±25 M€ and would be cost-effective if CSFV was introduced once every 2,000 years. Finally, leaving out *inspection* saved 12.5 M€ per year and did not increase epidemic costs. Since CSFV introductions into The Netherlands are expected every 2 - 18 years (Horst et al., 1999; De Vos et al., 2004), all programmes apart from *inspection* are very cost-effective. Notably, cost-effectiveness would increase if all costs were considered.

Unlike in previous studies (Crauwels et al., 1999; Stegeman et al., 2000), this paper shows a simultaneous analysis of surveillance programmes, enabling a comprehensive comparison between programmes. However, some model limitations need to be considered. The most important limitation of the epidemic and clinical disease models was that they were only based on data from the CSFV strain of the Dutch 1997-1998 CSF epidemic. Other strains might lead to other transmission parameters and above all, to other clinical disease parameters.

Important limitations of the surveillance programme models consisted of the starting day distributions, as no data were available. In the model, it was assumed that all herds with severely diseased pigs would sooner or later submit a pig for gross pathology, which would also lead to a tonsillar test for such herd. However, a 100% submission rate is not realistic, but determining the actual submission rate was impossible, as there exists no protocol for pig submission. Therefore, we assumed 100%, realising that the results will be too optimistic for both *pathology* and *tonsil*.

Deciding whether to continue with a surveillance programme cannot be based only on this study. Aside from ethical arguments, which cannot be assessed by calculations, some surveillance programmes have more objectives than just CSF surveillance: *inspection* is also used for FMD surveillance, and *pathology* for endemic disease diagnosis.

Nevertheless, the model does give an indication of the effectiveness and costs of CSF surveillance. The most important conclusions are that the current surveillance programmes can prevent expensive epidemics with high probability and that application of the four-weekly clinical inspection might need to be reconsidered.

ACKNOWLEDGEMENTS

We wish to thank Dr. A. Elbers (AHS), Dr. W. Loeffen (CIDC), S. Mesman (VWA), and H. van Langen (VWA) for their contribution to the description of the surveillance programme models. The research was supported by the Food and Consumer Product Safety Authority (VWA), The Hague, The Netherlands.

REFERENCES

Bouma, A., Stegeman, J.A., Engel, B., De Kluijver, E.P., Elbers, A.R.W. and De Jong, M.C.M. (2001). Evaluation of diagnostic tests for the detection of classical swine fever in the field without a gold standard. J. Vet. Diagn. Invest. 13, 383-388

Crauwels, A.P.P., Nielen, M., Stegeman, J.A., Elbers, A.R.W., Dijkhuizen, A.A. and Tielen, M.J.M. (1999). The effectiveness of routine serological surveillance: case study of the 1997 epidemic of classical swine fever in The Netherlands. Rev. Sci. Tech. Off. Int. Epiz. 18, 627-637

De Vos, C.J., Saatkamp, H.W., Nielen, M. and Huirne, R.B.M. (2004). Scenario tree modelling to analyse the probability of classical swine fever virus introduction into member states of the European Union. Risk Analysis (in press)

Elbers, A.R.W., Vos, J.H., Bouma, A., Van Exsel, A.C. and Stegeman, J.A. (2003). Assessment of the use of gross lesions at post-mortem to detect outbreaks of classical swine fever. Vet. Microbiol. 96, 345-356

Horst, S.H., Dijkhuizen, A.A., Huirne, R.B.M. and Meuwissen, M.P.M. (1999). Monte Carlo simulation of virus introduction into The Netherlands. Prev. Vet. Med. 41, 209-229.

Klinkenberg, D., Everts-van der Wind, A., Graat, E.A.M. and De Jong, M.C.M. (2003). Quantification of the effect of control strategies on classical swine fever epidemics. Math. Biosci. 186, 145-173

Mangen, M.-J.J. (2002). Economic welfare analysis of simulated control strategies for classical swine fever epidemics (PhD thesis). Wageningen Agricultural University, Wageningen, 188p

Mangen, M.-J.J., Nielen, M. and Burrell, A.M. (2002). Simulated effect of pig-population density on epidemic size and choice of control strategy for classical swine fever epidemics in The Netherlands. Prev. Vet. Med. 56, 141-163

Stegeman, J.A., Bouma, A., Elbers, A.R.W. and Verheijden, J.H.M. (2000). The white blood cell count is a valuable parameter for detection classical swine fever [Leukocytentelling is een waardevol hulpmiddel voor het opsporen van klassieke varkenspest]. Tijdschr. Diergeneesk. 125, 511-518

ENDEMIC DISEASES

UNDERSTANDING THE INFECTIOUS AETIOLOGY OF FOOTROT IN SHEEP: OPTIMISING INTERVENTIONS TO MINIMISE DISEASE

L.E. GREEN[1], G.J. WASSINK, R. GROGONO-THOMAS, L.J. MOORE AND G.F. MEDLEY

SUMMARY

A 'Best Practice' pilot leaflet to minimise footrot (FR) was drawn up and sent to farmers. Farmers were asked whether they would adopt the recommendations. Approximately 90% of farmers replied that they would adopt some of the recommendations. In 2003 a follow up study was conducted and these farmers were asked to estimate the level of footrot in their flock in 2002, what treatment and control methods they were using and when they first started these practices. Approximately 80% of farmers had been using the same control practices for at least five years. Those that routinely trimmed their sheep's feet reported a high prevalence of FR in 2002 and a dose response of more trimmings related to a higher prevalence of FR was still present. Individual treatment was still highly successful. From 2002-3 a cohort study monitored FR levels together with individual and flock control practices on 7 farms through visits and farmer data collection. Analysis of one of these farms where individual records were collected indicated that certain sheep and feet were more prone to footrot. Sheep that had had interdigital dermatitis were four times as likely to present with FR 1 – 2 months later. A binomial mixed effect model with presence of footrot or ID as the dependent variable indicated that the amount of FR and ID in the flock in the previous 4 and 6 weeks influenced the occurrence of FR and ID at any time interval. Routine foot trimming significantly increased the risk of FR and ID and treatment (parenteral antibiotic injection together with foot trimming) reduced the risk of FR and ID four weeks later. Trimming diseased feet without parenteral antibiotic increased the risk of FR/ID four weeks later. It was concluded that FR / ID are infectious diseases and that management must take this into account. Individual treatment of diseased sheep is recommended to control FR in the whole flock. Routine foot trimming should be avoided.

INTRODUCTION

Footrot (FR), caused by *Dichelobacter nodosus,* and interdigital dermatitis (ID), caused by *Fusobacterium necrophorum,* are the most important causes of lameness in sheep in the United Kingdom (Grogono-Thomas & Johnston 1997). The diseases have persisted at high levels for many decades despite concerted efforts for both treatment and control. In 1999 MAFF (now Defra) funded a research project (AW1007) to test the hypothesis that traditional techniques to manage FR in sheep in the UK were unsuccessful either because a) they did not work *per se* or because b) farmers were not able to carry out these methods to a high enough standard to achieve efficacy.

---

[1] Laura Green, Ecology and Epidemiology Group, Department of Biological Sciences, The University of Warwick, Coventry CV4 7AL. Email: laura.green@warwick.ac.uk

In 1999, the recommended techniques for managing FR in the flock were quarantine of new stock, routine trimming of feet, regular footbathing and vaccination and the recommended treatments for diseased sheep were isolation, parenteral antibiotics, topical foot spray and paring of the diseased foot (Morgan, 1987).

In 2000, 392 farmers who had participated in a previous study by Grogono-Thomas and Johnson (1997), and had said that they would participate in further research, were contacted by post. They were asked to complete a detailed questionnaire on levels of infectious lameness (footrot, interdigital dermatitis and contagious ovine digital dermatitis) in their sheep over a period of one year and their management practices to prevent and treat lameness in the same year. Results from this retrospective cross sectional postal survey (Wassink et al., 2003) of 209 farmers indicated that the isolation of new stock, and the separation and treatment of all clinically diseased sheep using all current recommendations was associated with a low prevalence of FR (2.6%). The more treatments a farmer used on all sheep the lower the prevalence of FR (3% c.f. 15%). However, control methods such as routine foot trimming, footbathing and vaccination were not associated with low levels of footrot. They were, at best, ineffective and, at worst, associated with a higher prevalence of FR. There was an apparent dose response effect of routine foot trimming: the more trimming events, the higher the prevalence of FR (6% for 0 –1 trims per year to 16% for > twice per year). These results indicated that individual treatment of FR was successful but that measures to control FR were ineffective and possibly counterproductive. However, the results from Wassink et al. (2003) may have occurred because of temporal error since management and disease were reported for the same time period.

We modified our hypothesis to: gathering sheep and routine foot trimming may enhance transmission through increased contact between sheep and successful transmission of *D. nodosus / F. necrophorum* on contact, whilst treating individual diseased sheep decreases the concentration of *D. nodosus* and therefore reduces transmission and disease. This hypothesis was explored by following individual and flock management of footrot and ID through time.

MATERIALS AND METHODS

1). A follow up of all 209 farmers

In 2001, a 'Best Practice' pilot leaflet, to minimise FR, was drawn up and is now available at http://template.warwick.ac.uk/staff/lgreen/Footrot/Practice.htm and sent to all the sheep farmers who had replied to our first study (Wassink et al., 2003). Farmers were asked whether they would adopt our recommendations. Approximately 90% of farmers replied that they would adopt some, but not all, of the recommendations. In 2003, 80 of these farmers were contacted again and asked to estimate the level of FR in their flock in 2002, what treatment and control methods they had used and when they first started these practices.

2) Longitudinal study

In 2002-3 a longitudinal study, monitoring the incidence of FR, together with individual treatment and flock control practices was set up. Seven farms participated and were visited every eight weeks for 15 months. Farmers were asked to collect data on the occurrence of footrot between visits. The method of analysis and results from one of these farms is presented below.

Data analysis: A binomial two-level model accounting for repeated examinations of sheep was set up with a two-week interval defining a new occurrence of FR or interdigital dermatitis (ID). The model was set up to estimate the lag in time from a management practice to the event of FR/ID in a sheep. This is described in the equation:

$$\text{Logit (FR/ID)}ij = \alpha ij + \Sigma\beta j + uj + eij$$

where FR/ID = the occurrence of footrot or interdigital dermatitis, α = intercept, β = coefficients, uj = between sheep variability and eij = residual error (fixed at mean 0, variance 1), i = examination, j = sheep

The $\log_{10}$ sum of the number of sheep with the FR, ID, treated for FR or ID (by injection or spray),foot trimmed because of footrot and foot trimmed as a routine procedure were estimated. These independent variables were lagged from two to eight weeks from the actual event. Housing was coded as a two level variable of not housed or housed for less than 3 months versus housed 3 – 6 months.

RESULTS

1). Follow up study

Fifty-four (68%) useful responses were received. Of these, 77% of farmers had not changed routine foot trimming practices and 81% had not changed foot-bathing procedures for more than five years. Fifty percent of farmers had started to treat individual sheep since 2002. Those that routinely trimmed their sheep's feet reported a higher prevalence of FR in 2002 and a dose response of more trimming events was again associated with a higher prevalence of FR when compared with farmers who did not trim the feet of their sheep. Individual treatment was still highly associated with low levels of footrot. Approximately 60% of farmers reported that the recommendations sent to them had resulted in lower levels of FR in 2002.

2). Longitudinal study

In the longitudinal study, there were 166 sheep – 20 sheep were omitted because of lack of identification. 97 (59%) sheep were affected with FR or ID at some point in time between February 2002 and July 2003. There were 33 FR cases/100 sheep/yr and 39 ID cases/100 sheep/yr. The average number of feet affected was 1.4 feet for FR and 2.1 for ID (Table B). If a foot had been treated for FR and/or ID on a previous occasion that same foot had an increased risk of being diseased for a second time of RR=1.91 (1.32 – 2.77) Chi2 = 12.7 $P<0.01$ (Sheep treated within 14 days of previous treatment were considered to be the same disease occurrence). The number of treatments ranged from one treatment in 24.4 sheep / 100 sheep / year to five treatments given to 0.5 sheep / 100 sheep / year.

The average number of days between treatments of the same sheep (>14 days was 165 days (range 16 – 410). Sheep that had interdigital dermatitis were four times more likely to present with FR 1 – 2 months later. Foot trimming significantly increased the risk of FR four weeks later and treatment using parenteral antibiotics reduced the occurrence of disease. The results from the mixed effects binomial model indicated that the occurrence of footrot or scald (FR/ID) in a sheep was predicted by the log 10 sum of FR and ID in the previous 4 and 6 weeks. FR/ID were positively associated with routine trimming of feet four weeks previously and with housing for more than three months. FR/ID was negatively associated with log 10 number of sheep treated with parenteral antibiotic 4 weeks previously and the number of diseased sheep whose feet were trimmed 4 weeks previously. The effect of spraying feet was non-significant although the trend was that it was protective.

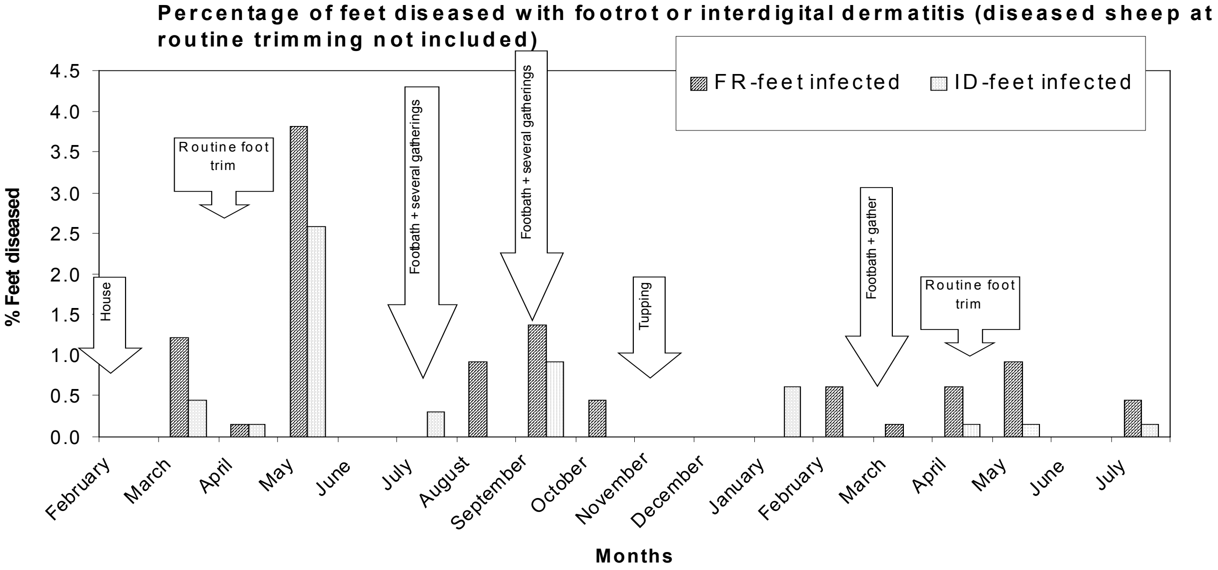


Fig. 1 Schematic diagram of the detection of footrot and interdigital dermatitis and the management of sheep on the farm

Table 1. Final binomial mixed effects model.

Variable	Coefficient	Standard error	Odds ratio	Lower 95 CI	Upper 95 CI
Intercept	-6.13				
Log10 sum sprayed to treat FR or ID 4 weeks previously	-0.58	0.45	0.56	0.23	1.38
Log10 sum FR 4 weeks previously	3.12	0.66	22.65	6.05	84.77
Log10 sum FR 6 weeks previously	-3.6	0.44	0.03	0.01	0.07
Log10 sum ID 4 weeks previously	0.53	0.67	1.70	0.44	6.49
Log10 sum ID 6 weeks previously	2.25	0.31	9.49	5.10	17.64
Log10 sum trimmed with FR 4 weeks previously	-2.66	1.24	0.07	0.01	0.84
Log10 sum routine trim 4 weeks previously	2.2	0.71	9.03	2.18	37.34
Housed for > 3 months	1.72	0.41	5.58	2.46	12.68
Log10 sum injected to treat FR 4 weeks previously	-10.28	1.03	3.43×10^{-5}	4.37×10^{-6}	2.69×10^{-4}

Random sheep effect =1.69 +/- 0.42

DISCUSSION

The data and results of statistical analysis are supportive of our modified hypothesis for this flock. Individual treatment of diseased sheep was associated with prevention of FR/ID whilst routine foot trimming was associated with FR/ID. The results are consistent with FR/ID as an infectious disease in which interventions to reduce susceptibility or transmission are effective means to control the incidence and prevalence of disease.

Running the model with FR alone gave similar results to those above but less power, because there were fewer events. We combined footrot and interdigital dermatitis because all the sheep with either presentation were lame and the farmer categorised the sheep as affected with FR or ID on clinical signs. Whilst there is a clinical difference between FR and ID (FR includes under running of hoof horn and damage to the interdigital skin whilst ID involves damage to the skin alone) we suggest that there is less evidence that these two clinical manifestations are bacteriologically distinct. They may be part of the same infectious process. Anecdotal evidence for this comes from observations that eliminating FR from a flock also eliminates lameness caused by ID. In addition, when Parsonson et al., (1967) first described ID they reported a transient clinical sign of abnormalty in the interdigital skin in which lameness was absent. Interdigital dermatitis with lameness has been thought to be caused by *F. necrophorum* alone. Our evidence (Moore et al., in preparation) indicates that over 60% of sheep with ID (and lame) had *D. nodosus* present, a similar proportion to those with FR. Further study of the aetiology of ID and FR may be beneficial.

A binomial model was selected to study this farm because we had individual sheep data over time and events that occurred in time. There was a clear effect of variability between sheep, indicating that certain sheep were apparently more susceptible to FR/ID.

The model was set up to estimate the lag in time from a management practice to the event of FR/ID in a sheep. These lags became significant 4 weeks after the independent variable had occurred. This suggests that disease may take more than 2 but less than 4 weeks to develop. The cyclical nature of disease (Fig. 1) was described by the occurrence of FR and ID in the previous four and six weeks and also by the treatments for FR and routine foot trimming. Calendar time was not significant but seasonal changes may have been having an effect on disease. However, results from Wassink et al., (2003) indicate that farmers do not identify a seasonal effect of FR across flocks.

An alternative approach would have been to use survival analysis and this will be explored in future work. Interactions between disease and individual treatments may also impact on the subsequent level of FR/ID and will be explored. The very high odds ratios are unlikely to be quantitatively accurate since they occur when events are highly associated with a rare outcome. The wide range in the confidence interval indicates the 95% range where the true odds ratio may fall.

Routine foot trimming is now considered inappropriate by many experts in the field of footrot control (VR, April 19, 2003, pp 510-511 and Sheep Farmer July/August, 2003, pp 22-23). However, it is not yet clear whether farmers have also stopped using foot trimming as a routine procedure. Many farmers do perceive that treatment of individual sheep is effective, irrespective of the level of footrot in the flock (Wassink et al., in press), but many do not treat all cases of FR.

We need to encourage farmers to adopt all of these practices for every affected sheep since when all of these treatments were given to every FR diseased sheep the prevalence of FR was very low at 2.6% (Wassink et al., 2003). This should be possible since there is no requirement to change farmers' perception of use of these treatments except to expand on their use.

ACKNOWLEDGEMENTS

The authors would like to thank the farmers for their contribution to this project. This study was funded by the Department for Environment, Food & Rural Affairs (Project No. AW1007).

REFERENCES

Grogono-Thomas, R. and Johnston, A.M. (1997). A study of ovine lameness. MAFF final report. MAFF open contract oc59 45k

Morgan, K.L. (1987). Footrot. In Practice, July 1987, 124-129

Parsonson, I.M., Egerton, J.R. and Roberts, D.S. (1967). Ovine interdigital dermatitis. J. Comp. Path. 77, 309

Wassink, G.J., Grogono-Thomas, R., Moore, L.J. and Green, L.E. (2003). Risk factors associated with foot rot in sheep Vet. Rec. 152, 351-358

Wassink, G.J., Grogono-Thomas, R., Moore, L.J. and Green, L.E. (2004). Risk factors associated with the prevalence of interdigital dermatitis (id, strip) in sheep in England and Wales over a one-year period. Vet Rec. In press

FACTORS ASSOCIATED WITH MILK QUALITY OF SMALLHOLDER DAIRY FARMS IN THE 10TH REGION OF CHILE

G. VAN SCHAIK[*], L.E. GREEN, D. GUZMÁN, H. ESPARZA AND N. TADICH

SUMMARY

A study was carried out to investigate the principal management factors that influenced bulk milk somatic cell count (BMSCC) and total bacterial count (TBC) of smallholder dairy farms in the 10th Region of Chile. One hundred and fifty smallholder milk producers were randomly selected from 42 Milk Collection Centres (MCCs) and a detailed questionnaire was conducted. In addition, the BMSCC and TBC results from the previous two months fortnightly tests were obtained from the MCCs and the mean was normalised by a natural logarithm (LN) transformation. Multivariable regression models were used to associate the mean BMSCC and TBC with the binary management variables (present (1) was compared with absent (0)). A random MCCs effect was included in the models to investigate the importance of clustering of herds within MCC.

The final model for mean LNTBC explained 35% of the total variance. In the null model the random effect of MCCs was highly significant. It was explained by three covariates: milk collected once a day or less compared with collection twice a day, not cleaning the bucket after milking mastitic cows versus cleaning the bucket and cooling milk in a vat of water versus not cooling milk or using ice or a bulk tank to cool milk. Other factors that increased the LNTBC were a waiting yard with a soil or gravel floor versus concrete, use of plastic buckets for milking instead of metal buckets, not feeding California Mastitis Test (CMT) positive milk to the calves and cows of dual purpose breed. From the model it was predicted that a herd that did not comply with any of these management factors had a predicted TBC of 59×10^9 colony forming units (cfu)/ml whereas a herd that complied with all the management practices had a mean predicted TBC of 105 cfu/ml.

The model of mean LNBMSCC explained 18% of the variance; the random effect of MCC was not significant. Management factors that decreased the mean LNBMSCC were: using the CMT for one year versus using the test for more than one year or not at all, absence of a concrete waiting yard, not filtering the milk or using filters other than a plastic sieve to filter the milk, milking cows with mastitis last, and sometimes or always examining the udder before milking. A herd that complied with all of these management factors had a BMSCC of approximately 46,166 cells/ml whereas a herd that did not comply with all management practices above had a mean BMSCC of 2×10^6 cells/ml.

---

[*] Gerdien van Schaik, Instituto de Medicina Preventiva, Facultad de Ciencias Veterinarias, Universidad Austral de Chile, Valdivia, Chile. Email: gvanschaik@uach.cl

INTRODUCTION

In a recent study, Tadich et al. (2003) targeted medium to large dairy cattle farms in Chile to investigate management factors that influenced bulk milk somatic cell count (BMSCC). On these medium to large dairy farms progress is rapid and husbandry mimics that adopted by the USA and Europe. A postal questionnaire was sent to 3710 dairy farms via the eleven milk-processing plants that they supplied. The response proportion from Tadich et al. (2003) was disappointing (14%) and indicated that alternative techniques to collect data in Chile might be more appropriate. We therefore chose to randomly select farms and to visit them to interview farmers.

Eighty percent of the Chilean dairy herds and 66% of the Chilean milk is produced in the 10th Region (INE, 1997). About 84% of the 11,000 dairy herds in the 10th Region are considered smallholder dairy farms. These are often subsistence farmers with 2 - 40 cattle that produce less than 100,000 kg of milk per year (Amtmann et al., 1995). The farmers frequently have other enterprises (beef calves, poultry, and vegetables), use family labour and live off their own produce. They sell milk to obtain a continuous flow of cash to support their family and the property can also serve as a guarantee to obtain credit from a bank (Amtmann et al., 1995). Milk is usually collected by hand into buckets and poured into churns or collected by machine directly into churns. The churns are then left at the farm entrance and collected by a local co-operative milk collection centre (MCC) once or twice per day. At the MCC the quality of the bulk milk from each farm (bulk milk somatic cell count (BMSCC)) and total bacterial count (TBC) is checked before it is added to milk collected from other farms and cooled in a large refrigerated tank. There are 42 MCC in the 10th Region of Chile with 2,200 producers delivering their milk to these MCCs. The association is voluntary but restricted by geographic location of the farm relative to the MCC. The number of producers for each MCC varies during the year, but on average a MCC has 56 producers delivering milk throughout the year (ACOLECHE, 2003, personal communication).

Whether milk is intended for the liquid market or for further processing its keeping and processing quality is reduced when there are high concentrations of bacteria and/or somatic cells in the milk (Ma et al., 2000). Bacteria in milk originate from three sources: the environment, intramammary infections and normal udder flora (Pankey, 1989). High bacterial counts in milk are commonly associated with improper sanitation or poor cooling of the milk (Pankey, 1989) whilst the BMSCC is an indicator of the bacterial infection level in the quarters (Wilson et al., 1997).

Good milk quality is important for a profitable dairy industry. The EU, New Zealand and Australia require that milk used for dairy products have BMSCC levels below 400,000 cells/ml, Canada below 500,000 cells/ml, and the USA below 750,000 cells/ml (Sargeant et al., 1998; Norman et al., 2000; Van Schaik et al., 2002). In terms of bacterial standards, regulatory penalties are typically paid on milk with TBC>100,000 bacteria/ml. Chile is keen to expand both its liquid milk and processed milk markets to other countries and so has introduced penalties to improve milk quality, these penalties are also applied to smallholder dairy farmers. It is therefore important for the prosperity of these farmers that they produce milk of the highest quality.

The purpose of this study was to investigate factors that influence milk quality of smallholder dairy farmers in 10th Region of Chile and to identify factors that could be changed to improve milk quality.

MATERIALS AND METHODS

Study data

One hundred and fifty smallholder dairy cattle farmers were randomly selected from 42 MCCs in the 10th Region of Chile using sampling proportional to size of MCC. The herds had to produce milk all year round to be included in the study. The questionnaire used by Tadich et al. (2003) was adapted to an interview format and adapted to include practises used by smallholder dairy farmers. The questionnaire was pilot tested on 5 farms by two interviewers (DG and HE). Further changes were made to the questionnaire and it was then used in its final form. In April and May of 2002 the two data collectors visited all 150 farms with the MCC veterinarian (when available). At each farm the data collectors interviewed the farmer and checked that there were consistencies between the farmers response and what was observed on the farm e.g. floor material, water supply, type of bucket used could be checked. Data were collected on 300 variables that were possible risks for BMSCC and TBC. The fortnightly tests on BMSCC and TBC in the two months prior to the survey were obtained for each farm from the relevant MCC. The BMSCC was estimated using Fossomatic^{TM} FC (Foss A/S, Denmark). The MCCs tested bacterial count in one of two ways, Bactoscan^{TM} FC (Foss A/S, Denmark) or reduction time of methylene blue.

Data analysis

The data were checked for errors and outliers and queries were cross checked with the raw data. All analyses were carried out in SPSS for Windows version 11.0.1 (SPSS Inc, 2001). Measures of central tendency and dispersion were used to investigate continuous variables (Table 1) and frequency distributions to investigate categorical variables. The mean BMSCC and mean TBC of the four observations in the two months prior to the visit were used as the dependent variables in the analyses. These variables were normalised by a natural logarithm transformation (LN). Normality was investigated by visual inspection of the histograms and the Kolmogorov-Smirnov test statistic. The association between BMSCC and TBC was determined with a Pearson correlation coefficient. All independent management variables were categorised into binary outcomes and present was compared with absent. The management factors with Pearson correlation coefficients with $P \leq 0.20$ were tested in two multiple linear regression models, one each for mean LNBMSCC and mean LNTBC as the dependent variables. Backward elimination was used to exclude non-significant ($P>0.05$) management factors from the saturated models. Variable were forced in the models to correct for herd size, yield and the effect of the two methods of bacterial counting (BactoScan^{TM} and reduction time of methylene blue).

Multivariable mixed regression models were used to correct for the possible clustering of herds within MCC. Variance components (VC) and compound symmetry (CS) covariance structures were considered and the covariance structure with the lowest Akaike's Information Criteria was included in the model when significant (Akaike, 1974).

The fit of the models was based on the explained variance, represented by the adjusted R^2, and checked by plotting the standardised predicted values against the standardised residuals. Outliers with large residuals (<-2 and >2) were excluded from the model to check the robustness of the models. Collinearity between the covariates was checked by calculating Pearson correlation coefficients for all covariates related to LNBMSCC or LNTBC with a $P \leq 0.10$ (Table 4 and 5). The Pearson correlation coefficient for two binary variables is equivalent to the phi correlation coefficient (Yule, 1912). A significant difference was assumed when *P*≤0.05 unless mentioned otherwise.

RESULTS

Descriptive statistics

From the 42 MCC, 1 to 10 farms were included in the study. The herds had on average 28 ha. of land and 11 milking cows that produced 7.3 kg of milk per cow per day. The mean rate of mastitis was approximately 17 cases per 100 cows per year.

The TBC was not determined by 7 MCCs, which excluded 30 herds. The TBC of 82 herds was based on BactoScan^{TM} and for the remaining 38 farms from 11 MCCs the reduction time of methylene blue was used. The mean TBC was 332,000 cfu/ml; 40% of the herds had a TBC below 100,000 in the four fortnightly observations. The mean TBC estimates were not significantly different by method of measurement. The geometric mean BMSCC was 408,000 cells/ml: 45% of the herds had a BMSCC < 400,000 cells/ml in the four fortnightly observations.

Table 1. Descriptive statistics of the smallholder dairy farms in the 10th Region of Chile.

	N	Mean	SD	Min.	Max.
Mean total bacterial count in bulk milk (1000 cfu/ml)	120	332	822	2.5	5,018
Mean bulk milk SCC (1000 cells/ml)	150	408	309	40.5	1,925
Farm size in ha.	149	28.0	21.6	2	100
Herd size	150	20.5	14.2	3	70
Number of milking cows	150	10.7	7.9	1	42
Average daily milk production (kg)	150	7.3	4.7	2.5	50
Rate of mastitis in the milking cows in the previous month (per 100 cows)	107	7.0	17.0	0	100
Yearly rate of mastitis in the milking cows (per 100 cows)	149	16.8	21.3	0	150

The LNTBC did not follow a perfectly normal distribution. For BMSCC both the Kolmogorov-Smirnov test statistic (*P*>0.20) and the histogram indicated that the LNBMSCC was normally distributed.

Multivariable model for LNTBC without random effect for MCC

The final linear regression model for LNTBC is presented in Table 2. When the null model for TBC was investigated the random effect for MCC with a VC covariance structure had a strong effect (intraclass correlation MCC=41%, *P*=0.02), indicating that a large proportion of the variability in TBC occurred at the co-operative level. When the independent variables were fitted and the model finalised, the effect of co-operative was explained by the variables labelled (*) in Table 2: these were frequency of milk collection by the co-operative, method that farmer used to cool their own milk, and washing the collecting bucket after milking a mastitic cow. The other variables that affected TBC are listed in Table 2. The final linear regression model had an adjusted R^2 of 35%. The residual plot showed a random distribution of the standardized residuals around zero. There were eight outlying observations but exclusion of these eight herds from the model did not change the estimates of the variables.

Table 2. Linear regression model for the natural logarithm of the total bacterial count (LNTBC) of 120 smallholder dairy farms in Chile.

	LNTBC	Std. Error	P	Farms with factor (%)	Change in LNTBC ($x10^3$)
Intercept	11.27	0.37	<0.01		
Milk is collected twice a day vs. once a day or less*	-1.34	0.69	0.05	5	-58
Metal bucket for milking vs. plastic buckets	-1.11	0.27	<0.01	65	-53
Wash the bucket after milking an mastitic cow vs. not washing it*	-0.92	0.27	<0.01	53	-47
Cooling the milk in a vat filled with water vs. not cooling it or use other methods for cooling*	0.86	0.29	<0.01	30	+107
A waiting yard with soil or gravel floor vs. a cement floor or no waiting yard	0.84	0.27	<0.01	65	+103
Dual purpose vs. dairy or other breed	0.82	0.27	<0.01	45	+178
Feeding CMT positive milk to the calves vs. not using CMT or combine the positive milk with the bulk milk	-0.73	0.32	0.02	22	-41

A farm that complied with all covariates in the above model had a predicted TBC of 105 cfu/ml whilst a farm that had all the risks for raised TBC had a predicted count of $59x10^9$ cfu/ml.

Multivariable model for LNBMSCC

The random MCC effect with a VC or symmetrical covariance structure did not explain significant variation in the null or final model (intraclass correlation coefficient=0.02, *P*=0.71). So a random effect for MCC was not included. The final model explained 21% of the variation in BMSCC (adjusted R^2=18%) and the model fitted the data well. Only four herds had standardized residuals <-2 or >2 and when these herds were excluded from the model the variable ‘Filter milk with a plastic sieve’ became non-significant at *P*=0.07.

A farm that practised no factors that increased BMSCC from the model has a predicted BMSCC of 46,166 cells/ml, whilst a farm with all the risk factors has a predicted BMSCC of almost 2,000,000 cells/ml.

Table 3. Linear regression model for the natural logarithm of the bulk milk somatic cell count (LNBMSCC) of 150 smallholder dairy farms in Chile.

	LNBMSCC	Std. Error	P	Farms with factor (%)	Change in BMSCC ($x10^3$)
Intercept	12.61	0.13	<0.01		
Use CMT for one year vs. no CMT use or use CMT for more than one year	-0.50	0.18	0.01	10	-118
Waiting yard with concrete floor vs. soil or gravel floor	0.45	0.15	<0.01	20	+170
Filter milk with a plastic sieve vs. not filtering or use of a metal sieve	0.32	0.12	0.01	34	+113
Milk mastitic cows last vs. in no specific order	-0.31	0.12	0.01	66	-80
Never examine the udder before milking vs. sometimes or always	0.29	0.12	0.01	40	+101

DISCUSSION

The design of this study was highly successful: all 150 randomly selected farms co-operated. Assuming that there was no unknown bias, the random study design with sampling of farms according to proportion within each co-operative gave accurate estimates of changes in BMSCC and TBC for smallholder dairy farms in the 10th Region of Chile. Farms had varying numbers of cows, milk production levels, and mastitis incidence and these were forced in the models but were not significant covariates.

The selection process using farms within co-operatives enabled examination of the source of the variability for each of the two dependent variables, BMSCC and TBC. In the TBC model, three covariates explained the clustering of farms within co-operatives. The most important of these was the number of milk collections per day: when co-operatives collected milk twice a day versus once or less than once per day the TBC was reduced by 58,000 bacteria/ml. This is a considerable reduction and indicates that milk quality was affected by number of milk collections per day, something outside the farmer's control. When milk is collected once a day, TBC can increase by environmental contamination (e.g. dust) and, especially during spring and summer, by high temperatures. Therefore, farmers try to keep the milk as cool as possible by introducing the milk churns into (running) water. Where farmers attempted to cool milk in a vat of water (30 herds) the TBC was on average higher than when the milk was not cooled (28 herds) or cooled using a proper cooling tank (2 herds) or bottles filled with ice (90 herds). The effect is probably because the water itself is likely to have high TBC, since farmers usually used local supplies of water or surface water, rather than mains water and thus the water can contaminate the milk. The third factor linked to co-operative was washing the bucket after collecting mastitic milk. This is clearly to be recommended to reduce bacterial contamination of milk and we think that this factor was associated with co-operatives because this practice was recommended by some but not all of the advisers of the MCCs.

The other four variables in the TBC model were not linked to MCC. The practice of not putting high SCC milk into the bulk milk (identified with CMT and feeding it to calves) is a sensible strategy to reduce TBC. Whilst it is obvious that metal buckets can be cleaned more thoroughly and dry faster than plastic buckets, they are more expensive; around US$10 for a metal bucket versus US$2 for a plastic bucket. The investment may be worthwhile for the 35% of the farmers that did not use metal buckets when it will decrease their TBC with 53.000 cfu/ml. Having a yard where cows stood before they were milked with a floor of soil or gravel compared with concrete was linked to high TBC. This may be because cleaning these yards is difficult (only 28% of these yards were cleaned versus 82% of the concrete yards) and they turn into mud pools during winter, which causes contamination of the udder. However, a concrete waiting yard may not be economically feasible for smallholder dairy farms. A more feasible option may be proper preparation of the udder such as, washing the udder and drying the teats before milking, which has been demonstrated to reduce TBC by 40%, particularly during winter (McKinnon et al., 1990). In our study, 84% of the farmers said that they washed the teats or udders of all cows before milking but this had a positive correlation with TBC. Of the farmers that wash the udders, 35% used water with disinfectant, 32% used a hose and 66% of them dried the teats after cleaning. The positive correlation between cleaning the udder and TBC indicated that a proper washing and drying routine should be emphasized. Finally, herds with a dual purpose breed had higher TBC than herds with a dairy breed. Part of that effect may be a true breed difference but the two breeds did not differ in milk production or mastitis incidence. Another explanation is that farmers with the dual purpose breed pay less attention to milk hygiene. The correlations showed that farmers with the dual purpose breed are less likely to wash the bucket after milking a mastitic cow. Finally, the BMSCC was not significantly correlated with TBC, indicating that the TBC were affected by improper sanitation or poor cooling of the milk and not by mastitic cows with high bacterial loads in their milk.

The factors in the final model explained 35% of the variability observed on TBC between farms. This is a good level of explained variability for an observational study. Whilst the dependent variable was not visually precisely normally distributed once logged, the standardised residuals indicate that the model was a good fit since these were Normally distributed.

Herds with BMSCC < 250,000 cells/ml are approaching the optimal level of udder health (Peeler et al., 2002), while herds with BMSCC > 500,000 cells/ml have significant problems with subclinical mastitis (Dohoo & Meek, 1982). The mean BMSCC of the study herds of 408,000 cells/ml is a moderate level that leaves room for improvement. The majority of the variability in BMSCC occurred on farms. As with Tadich et al. (2003) never examining the udder before milking versus sometimes or always examining the udder was associated with higher BMSCC. In contrast, in this study, milking mastitic cows last versus in no specific order was associated with reduced BMSCC, in Tadich et al. (2003) milking mastitic cows first was linked with low BMSCC. We think the current study findings are biologically more plausible since this should minimize contamination (Guterbock, 1984; Hutton et al., 1991; Wilson et al., 1996). CMT is a simple and cheap cow-side screening test to detect subclinical mastitis that gives an indication of the grade of infection of each quarter in cows (Blowey & Edmonson, 1995; Dingwell et al., 2003). Interestingly, using CMT for one year versus not using CMT or using it for more than one year was associated with lower BMSCC. This may indicate that those who recently started using CMT are more efficient at keeping CMT positive milk out of the bulk milk or it may be that they are using CMT to lower BMSCC and so have started to use the technique. Others who had used CMT for longer than one year may use it to perform the recommendations of the MCC's advisers but they were not interpreting the CMT reactions appropriately or were not taking the right measures such as discarding the milk or not sending the milk to the MCC. The data showed that 87% of the farmers that used CMT for one year used it before milking compared with only 46% of the farmers that used CMT for longer than a year, In addition, 40% of the one-year CMT users applied it once in 15 days compared with 10% of the long-term CMT users. Farmers who filtered milk with a plastic sieve versus not filtering or with a metal sieve were smaller herds (3 milking cows less than herds that did not use a plastic sieve) and had higher BMSCC. Low BMSCC has been associated with herds with better hygiene when compared with herds with high BMSCC (Barkema et al., 1998, Barkema et al., 1999). Thus, a possible explanation is that the producers with the plastic sieve had worse hygiene during milking and therefore had to filter the milk. Moreover, the effect disappeared when four outlying farms were excluded from the dataset, which indicated that the effect was merely due to these four poorly managed farms with high BMSCC. Farms with a waiting yard with concrete floor versus soil or gravel floor had higher BMSCC. This is in contrast to the impact of this floor type on TBC, where high TBC was associated with soil and gravel yards. It may be that the latter floor types reduce fecal contamination and the risk of udder infection but increase overall soiling, so the bacterial species would be different. Moreover, the interviewers (DG and HE) reported that some farmers with concrete yards kept their cows overnight in the yards, which may have enhanced environmental mastitis. The data showed a slightly higher but non-significant mastitis rate (~5% higher, P=0.21) in the herds with a concrete yard, which may explain the increased BMSCC. Keeping cows overnight in the yard should be included in the next version of a questionnaire as a potential risk factor. Other studies showed that collecting cows in the yard before milking increased the risk of BMSCC > 200,000 (Tadich et al., 2003) and that this increase was probably due to the cleanliness of the gathering yard; dirty yards have been associated with high BMSCC (Schukken et al., 1990; Wilson et al., 1997; Peeler et al., 2000). Farms with concrete waiting yards had significantly more cows and hectares of land (+5 cows and +10 ha.) than the other herds but did not differ in daily milk production.

Overall, the model for reducing TBC was more informative and biologically plausible than the BMSCC model. This is not surprising since it is primarily a measure of contamination whilst BMSCC is a measure of infection and so cow factors not measured in this study would have a large effect. This study has demonstrated that a considerable gain in milk quality and therefore profitability could be obtained if smallholder dairy farms in Chile complied with management practices reported in this study and when milk was collected from farms twice a day. Some of the recommendations such as use of metal buckets for milking, always examine the udder before milking, milk mastitic cows last, wash the bucket after milking a mastitic cow, use of CMT and discard CMT positive milk could be applied immediately and are not only applicable for the Chilean dairy herds but are universal recommendations.

ACKNOWLEDGEMENTS

The authors are very grateful for the assistance of the ACOLECHE, MCCs, the veterinarians and the dairy farmers for co-operating in the study.

REFERENCES

Akaike, H. (1974). A new look at the statistical model identification. IEEE Transaction on Automatic Control AC-19, 716-723.

Amtmann, C., Mujica, F. and Vera, B. (1995). Producción de Leche. In pequeña Agricultura en la Región de los Lagos, Chile. Valdivia, Chile, pp 75-91.

Barkema, H.W., Schukken, Y.H., Lam, T.J.G.M., Beiboer, M.L., Wilmink, H., Benedictus, G. and Brand, A. (1998). Incidence of clinical mastitis in dairy herds grouped in three categories by bulk milk somatic cell counts. J. Dairy Sci. 81, 411-419

Barkema, H.W., Schukken, Y.H., Lam, T.J.G.M., Beiboer, M.L., Benedictus, G. and Brand, A. (1999). Management practices associated with the incidence rate of clinical mastitis. J. Dairy Sci. 82, 1643-1654

Blowey, R.W. and Edmonson, P. (1995). Mastitis Control in Dairy Herds - An Illustrated and Practical Guide. 1st. ed. Farming Press, Ipswich, UK

Blowey, R., Edmondson, P. and Davis, J. (1999). Bacterial counts in bulk milk-an Update. In Practice 32, 531-534

Dingwell, R.T., Leslie, K.E., Schukken, Y.H., Sargeant, J.M. and Timms L.L. (2003). Evaluation of the California mastitis test to detect an intramammary infection with a major pathogen in early lactation dairy cows. Can. Vet. J. 44, 413-416

Dohoo, I.R and Meek, A.H. (1982). Somatic cell counts in bovine milk. Can. Vet. J. 23, 119-125

Guterbock, W.M. (1984). Practical aspects of mastitis control in large dairy herds. Part I. Assessing the status of Mastitis. Comp. Cont. Ed. 16, S601-S605.

Hutton, C.T., Fox, L.K. and Hancock, D.D. (1991). Risk factors associated with herd-group milk somatic cell count and prevalence of coagulase-positive staphylococcal intramammary infections. Prev. Vet. Med. 11, 25-35

INE (1997). VI Censo Nacional Agropecuario, Instituto Nacional de Estadística, Ministerio de Economía, Chile

Ma, Y., Ryan, C., Barbano, D.M., Galton, D.M., Rudan, M.A. and Boor, K.J. (2000). Effects of somatic cell count on quality and shelf-life of pasteurized fluid milk. J. Dairy Sci. 83, 264-274

McKinnon, C.H., Rowlands, J. and Bramley, J. (1990). The effect of udder preparation begore milking and contamination from the milking plant on bacterial numbaers in bulk milk of eight dairy herds. J. Dairy Res., 57 307-318

Norman, H.D., Miller, R.H., Wright, J.R. and Wiggans, C.R. (2000). Herd and state means for somatic cell count from dairy herd improvement. J. Dairy Sci. 83, 2782-2788

Pankey, J.W. (1989) Premilking udder hygiene. J. Dairy Sci. 72, 1308-1312

Peeler, E.J., Green, M.J., Fitzpatrick, J.L., Morgan, K.L. and Green, L.E. (2000). Risk factors associated with clinical mastitis in low somatic cell count British dairy herds. J. Dairy Sci. 83, 2464 - 2472

Sargeant, J.M., Schukken, Y.H. and Leslie, K.E. (1998). Ontario bulk milk somatic cell count reduction program, progress and outlook. J. Dairy Sci. 81, 1545-1554

Sargeant, J.M., Leslie, K.E., Shirley, J.E., Pulkrabek, B.J. and Lim, G.H. (2001). Sensitivity and specificity of somatic cell count and California mastitis test for identifying intramammary infection in early lactation. J. Dairy Sci. 84, 2018-2024

Schukken, Y.H., Grommers, F., van de Geer, D., Erb, H. and Bran, A. (1990). Risk factors for clinical mastitis in herds with a low bulk milk somatic cell count. J. Dairy Sci. 73, 3463-3471

Tadich, N., Kruze, J., Locher, G., Green, L.E. (2003). Risk factors associated with BMSCC greater than 200 000 cells/ml in dairy herds in southern Chile. Prev. Vet. Med. 58, 15-24

Van Schaik, G., Lotem, M. and Schukken, Y.H. (2002). Milk quality in New York State. Trends in Somatic Cell Counts, Bacterial Counts, and Antibiotic Residue Violations in New York State in 1999-2000. J. Dairy Sci. 85, 782-789

Wilson, D.J., González, R. and Sears, P. (1996). Segregation or use of separate milking units for cows infected with *Staphylococcus aureus* reduced prevalence of infection and bulk tank somatic cell counts. The Bovine Pract. 32, 40-42

Wilson, D.J., Das, H.H., Gonzalez, R.N. and Sears, P.M. (1997). Association between management practices, dairy herd characteristics, and somatic cell count of bulk tank milk. J. Am. Vet. Med. Assoc. 210, 1499 - 1502

Yule, G.U. (1912). On the methods of measuring association between two attributes. J. Royal Stat. Soc. 75, 579-652

RISK FACTORS FOR BECOMING A PERSISTENT CARRIER OF *SALMONELLA* DUBLIN IN INFECTED DAIRY HERDS

L.R. NIELSEN[1] AND A.K. ERSBØLL

SUMMARY

Salmonella enterica subsp. *enterica* serovar Dublin (*Salmonella* Dublin) is of concern in the cattle industry due to welfare problems and economic losses in infected herds and due to a serious zoonotic hazard to humans. Approximately 25% of Danish dairy herds are expected to be infected with the bacteria. Therefore, control of the infection is highly desired. *Salmonella* Dublin has a tendency to produce asymptomatic, persistent carriers in cattle, and these carriers are important for the spread of infection. Therefore, an important part of control strategies involve test-and-cull procedures of carriers, and there is a need for new knowledge regarding prevention of production of new carriers in infected herds. Results of the present study suggest that cattle that become infected with *Salmonella* Dublin as heifers (above 1 year old to 70 days before first calving) and cattle that become infected close to first calving or during first parity have the highest risk of becoming persistent carriers upon infection. Also, low herd prevalence of *Salmonella* Dublin leads to higher risk of becoming persistent carriers upon infection than high herd prevalence. Finally, significant variation was found between herds.

INTRODUCTION

Salmonella enterica subsp. *enterica* serovar Dublin (*Salmonella* Dublin) causes acute and chronic disease in cattle (Wray & Snoyenbos, 1985; Rings, 1985). It can cause disease in all ages of cattle though most commonly the clinically affected animals in infected herds are young calves (Wray & Davies, 2000). An important characteristic of *Salmonella* Dublin is the tendency of this bacteria to produce persistent carrier animals that show no symptoms but periodically shed large amounts of bacteria to the environment through faecal matter and milk and thus contribute to the spread of infection in the herds (Richardson, 1973; Spier et al., 1991; House et al., 1993). This property is assumed to be one of the major reasons that *Salmonella* Dublin, unlike other salmonella serotypes, often persist in cattle herds for decades with periodical disease outbreaks. *Salmonella* Dublin leads to both welfare and economical problems in such endemically infected herds. Common symptoms include diarrhoea, pneumonia, septicaemia, septic arthritis, loss of appetite and retarded growth in affected calves. Severity of the clinical symptoms depends on host and agent related factors such as age of the animal, virulence of the bacterial strain and number of bacteria ingested by the host. Abortion, diarrhoea and fever are the most frequently encountered symptoms in acutely infected adult cattle. Often, the symptoms

---

[1] Liza Rosenbaum Nielsen, Department of Animal Science and Animal Health, The Royal Veterinary and Agricultural University, Groennegaardsvej 8, DK-1870 Frederiksberg C, Denmark. E-mail: lrn@kvl.dk

will be so subtle that the infection goes unnoticed by the caretaker, but the animal may still become a persistent carrier of the bacteria and thereby become a risk for spread of infection.

Further, *Salmonella* Dublin is a serious zoonosis. Humans may become infected by eating contaminated undercooked beef meat or unpasteurised milk products. *Salmonella* Dublin has been shown to have a higher case fatality risk than other salmonella serotypes, e.g. *Salmonella* Typhimurium, most likely due to the invasiveness of the bacteria in the infected person (Fierer, 1983; Helms et al., 2003).

According to the Danish cattle salmonella surveillance program, approximately 25% of the Danish dairy herds are infected with salmonella (Nielsen et al., 2003a). Most of these are *Salmonella* Dublin infections. Due to the welfare, economic and zoonotic hazards that *Salmonella* Dublin poses, it is desirable to control the infection. The persistent carriers are important in keeping the infection present in the herds. Therefore, it is necessary not only to detect and cull such carriers, but also to avoid production of new persistent carriers. Hence, knowledge about what causes animals to become persistent carriers as opposed to clearing the bacteria from the organism upon infection with *Salmonella* Dublin is needed. House et al. (1991) reported that neonatal *Salmonella* Dublin septicaemia in calves may lead to systemic and mammary gland carriers of the bacteria when they become heifers. Spier et al. (1991) found that corticosteroid injections 7 weeks after inoculation of *Salmonella* Dublin into the mammary gland lead to recrudescence of bacterial excretion and lymphatic spread of the bacteria to the regional lymph nodes, suggesting that immunosuppression may play a role in the production of persistent carriers. Otherwise, little is known about why some animals become persistent carriers when they become infected with *Salmonella* Dublin, when others do not.

The objective of the present study is to examine host and environment related risk factors for becoming a persistent carrier upon infection with *Salmonella* Dublin in 12 infected cattle herds. A case-control study design is used for the analysis in which animals are categorized as either carriers or transiently infected based on repeated sampling.

MATERIALS AND METHODS

The present study was part of a large project known as “The Integrated Cattle Health and Milk Quality Project”, which was initiated by the Danish Dairy Board in 1997 (Andersen et al., 2000). Out of 249 dairy herds in the project region known as the Kongeå-region in the southern part of Jutland, 111 herds participated on a voluntary basis in projects about infectious diseases. Twelve of these herds (10 Holstein-Friesian breed and 2 Jersey breed) were selected to participate in the present study because they were known to be infected with *Salmonella* Dublin to an extend where it occasionally caused clinical problems in the herds. They also had to be willing to allow fairly extensive sampling in their herds. The main part of the sample collection consisted of 5 visits to each herd with approximately 3 months between visits. At each farm visit, rectal faecal samples were collected from all animals, heparin stabilized blood samples were collected from all non-lactating animals and milk samples were collected from all lactating animals on the premises. Six of the 12 herds also participated in a related project involving more frequent and intensive sampling of cows and calves after time of calving and birth. Monthly milk samples were collected from all cows through a milk quality recording scheme from Fall 2001 to Spring 2003 from these 6 herds.

All faecal samples were cultured for presence of salmonella bacteria at the Veterinary Department of Steins Laboratory in Ladelund, Brørup. The method has been described and the validity evaluated in previous studies. The sensitivity of the faecal culture method has been estimated to be between 6% and 32% (Nielsen, 2003) depending on the age of the animal and whether pooling of samples was used before individual follow-up. The specificity is assumed to be 100%, because typing of all salmonella positive isolates was performed at the Danish Veterinary Institute in Copenhagen. Blood samples and milk samples were analysed for presence of antibodies directed against *Salmonella* Dublin lipopolysaccharides (LPS) using an enzyme-linked immunosorbent assay (ELISA) that has been described in detail elsewhere (Hoorfar et al., 1994; Nielsen, 2003). An ODC%-value, which is a background corrected ratio of the test sample optical density (OD) to a known positive reference sample, was calculated for each sample as follows:

$$ODC\% = \frac{(\overline{OD}_{sample} - \overline{OD}_{neg\ ref})}{(\overline{OD}_{pos\ ref} - \overline{OD}_{neg\ ref})} * 100\%$$

where $\overline{OD}_{sample}$ is the mean value of two test wells, $\overline{OD}_{neg\,ref}$ and $\overline{OD}_{pos\ ref}$ are the mean values of four reference wells in the ELISA plates, respectively. The test sensitivity of the milk ELISA was estimated to be 50% (95% CI: 33-67%) and the specificity 93% (95% CI: 91-95%) for every sampling event at a cut-off value of 48 ODC%. For the serum ELISA, the cut-off value used in the present study was 35 ODC%, which gave a sensitivity of 65% (95% CI: 55-75%) and a specificity of 97% (95% CI: 96-98%) for every sampling event. If an animal seroconverted between two sampling dates, the sample immediately after seroconversion was denoted positive even if the cut-off value of 35 or 48 ODC% had not been reached yet. Seroconversion was defined as a rise in ODC% of more than 25 in the serum ELISA and more than 30 in the milk ELISA. Since most animals were tested using both one ELISA and one bacterial culture test, parallel sensitivities and specificities were calculated, and these parallel validity estimates were used to adjust prevalence estimates in the herd to obtain true herd prevalence estimates (TP) as described by Martin et al. (1987) for each quarter of the year.

In the 12 study herds, a total of 3277 animals were tested at least once during the study period, but in order to be included in the study, the animals had to be tested at least 3 times over a period of at least 270 days. No more than 180 days were allowed between two samples. The animals had to be laboratory test negative (below cut-off 35 (serum) or 48 (milk) and bacterial culture negative) at the first sampling event, and the infection date was estimated to be halfway between the first possible infection date (set to 7 days prior to the last negative test result), and the last possible infection date (set to 14 days prior to the first positive test result). However, for calves under the age of 1 year, if the first sampling event was positive, the animal was still included in the study and the infection date was estimated to be half way between birth and the first sampling date. Animals were categorised as persistent carriers (cases) and transiently infected (controls) according to the following criteria:

Carriers: At least 2 test positive sampling events with more than 270 days between the first and the last positive sampling event, and with the last sampling event having a positive laboratory test result. Persistent carriers are expected to have a continuous stimulation of the humoral immune system from the bacteria located in the host tissues, and therefore ODC%-values had to be above 60 in the milk ELISA and above 50 ODC% in the serum ELISA in order to be considered positive for the carrier categorisation. These criteria lead to a total of 161 carriers in the study from the 12 study herds.

Transiently infected: At least 1 test positive sampling event. Test positive sampling events for less than 150 days in total. The last sampling event had to have a negative laboratory test result. These criteria lead to a total of 104 transiently infected animals in the study from the 12 study herds. Examples of animals categorised as transiently infected and persistent carriers are shown in Fig. 1 and Fig. 2

Calving and production data were obtained from the Danish Cattle Database. Time from calving was calculated as the number of days between the estimated infection date and the last calving date, unless the animal had not calved before, in which case it would be a negative number indicating number of days before 1st calving. If a cow was less than 70 days before the next calving at time of infection, time from calving was calculated as time to next calving. A new categorical variable was constructed grouping the animals into 6 groups according to age and time from calving as illustrated in Table 1.

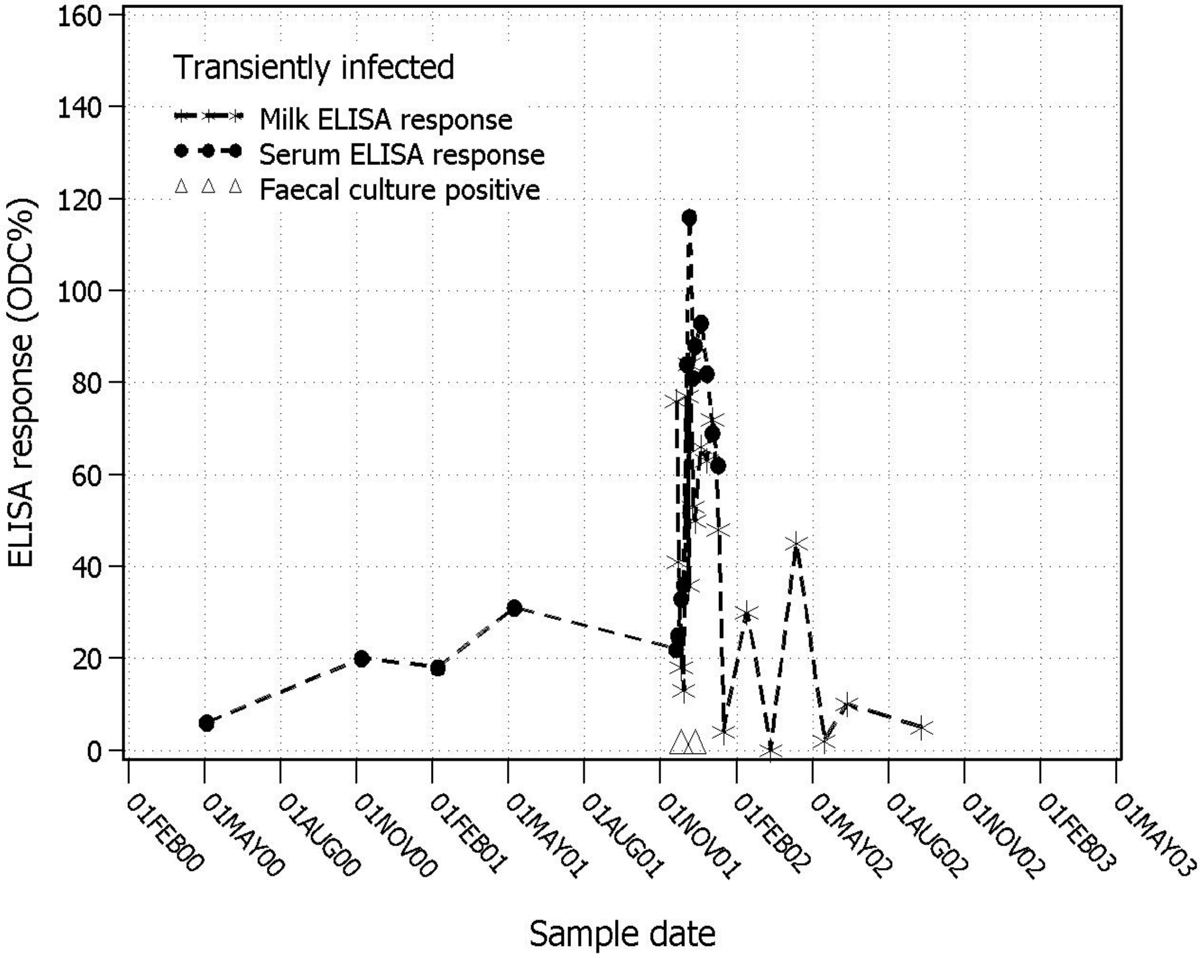


Fig. 1 Example of categorisation of a cow as transiently infected with *Salmonella* Dublin according to laboratory test results from bacterial culture of faecal samples and ELISA response in serum and milk samples.

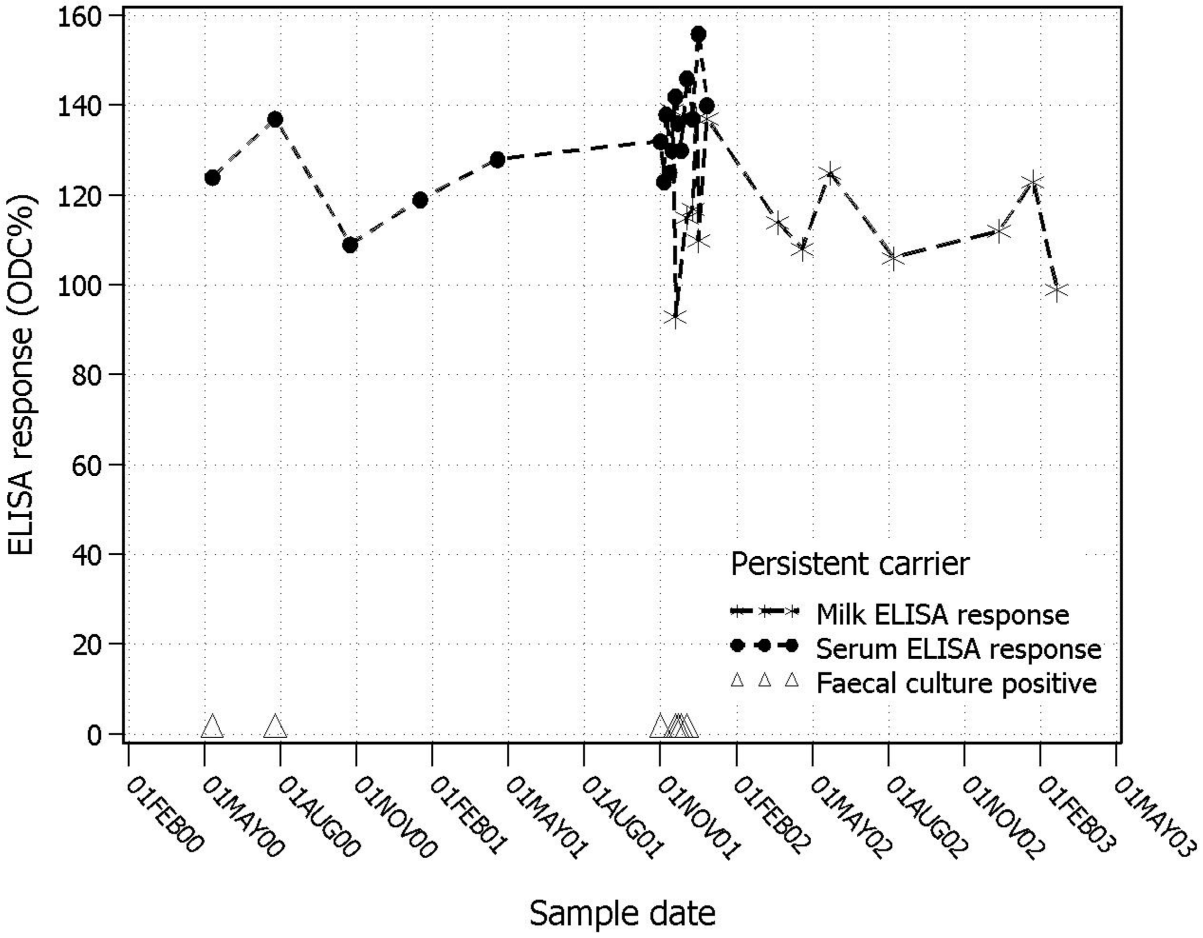


Fig. 2 Example of categorisation of a cow as persistent carrier of *Salmonella* Dublin according to laboratory test results from bacterial culture of faecal samples and ELISA response in serum and milk samples.

Table 1. Distribution of persistent carriers and transiently infected animals in 6 groups based on age and time from calving.

AGE AND TIME FROM CALVING GROUP	INFECTION STATUS			
	Transiently infected		Persistent carrier	
	n	%	n	%
Calves 0-1 year old	26	25.0	32	19.9
Heifers 1 year to 70 days before 1st calving	1	1.0	33	20.5
Heifers and cows ±70 days from 1st calving	9	8.7	19	11.8
1st parity cows in mid to late lactation	20	19.2	41	25.5
Cows 2nd or higher parity ±70 days from calving	5	4.8	5	3.1
Cows 2nd or higher parity in mid to late lactation	43	41.4	31	19.3
In total:	104	100	161	100

A multilevel, multivariable logistic regression model was built using backward elimination. To adjust for intra herd correlation, herd was included as a random effect in the model. The binary outcome in the model was carrier (1) or transiently infected (0). Risk factors tested in the model were all related to the estimated infection date of the individual animals in the study. The tested risk factors were: Age and time from calving group, season (quarter of the year), herd prevalence and all two-way interactions. Estimation of the different parameters was performed using the GLIMMIX macro in SAS® version 8.2. The deviance was evaluated for goodness-of-fit in relation to the degrees of freedom in the model. The fit of the final model was checked using Pearsons' residual plots for the full model and each variable (Collett, 1991).

RESULTS

The herd prevalence estimates showed a large variation between herds and time of sampling ranging from 0% to 84.2%. Below, it is graphically illustrated how the estimated true herd prevalence varied over time in herds that were sampled for 1-1½ years and herds that were followed for 2-3 years (Fig. 3). All herds except for one (herd number 2) were already infected before study onset. Some of the herds experienced outbreaks of clinical disease during the study period which usually lead to increases in the prevalence of infected animals.

All animals were categorised in different infection groups based on the results from the repeated sampling in the herds. Ideally, all animals could be categorised as either negative during the whole study period, transiently infected or persistent carriers. However, some animals were sampled too few times to be included in any of these infection groups, for instance animals that entered the herd late in the study or left the herd before the study was finished. Other animals had to be left out of the analysis, because they had positive laboratory test results between 150 and 270 days, i.e. between the criteria for being categorised as transiently infected or persistent carriers. Finally, animals that were older than 365 days and had a positive first sampling event were excluded from the analysis, because it was not possible to estimate the infection date with reasonable precision. Figure 4 shows the distribution of animals in five different infection groups according to the laboratory results from each of the 12 herds. Note that there is large variation in the proportion of the herds (39%-86%) that were sampled enough to be assigned to an infection group. There is also large variation between herds with regard to the proportion of persistent carriers according to the criteria used in the study. Varying proportions of the tested animals (20%-59%) had been infected, though it was not possible to tell, if they were carriers or transiently infected with the available data and the set criteria.

The following risk factors were found to be significantly associated with the risk of becoming a persistent carrier upon *Salmonella* Dublin infection in the model: Age and time from calving group ($p=0.02$), estimated true herd prevalence ($p=0.009$) and herd ($p=0.04$) which was included as a random effect. No interactions were found significant on a 20% significance level. The results from the final model are summed up in Table 2. Residual plots showed a few outliers but no trend in the plots. The model was tested without the outliers, and this did not change parameter estimates or statistical inferences made from the model to any noteworthy extent. Together, these finding suggest the model was well fit. The predicted risks for becoming carrier animals upon infection with *Salmonella* Dublin in the six age and time from calving groups are shown in Fig. 5. The resulting model was found to be as follows:

$$Logit(p_{ijk}) = \mu + AT_i + \beta * TP_{jk} + H_j + \varepsilon_{ijk}$$

where
p_{ijk} is the probability for becoming a persistent carrier for cow k infected in age and time group i in herd j.
μ is the general mean (the intercept)
AT_i is the fixed effect of age and time from calving, i=1….6
β is the slope
TP_{jk} is the true prevalence estimate in herd j at estimated infection time for cow k
H_j is the random effect of herd, j=1….12
ε_{ijk} is the random error

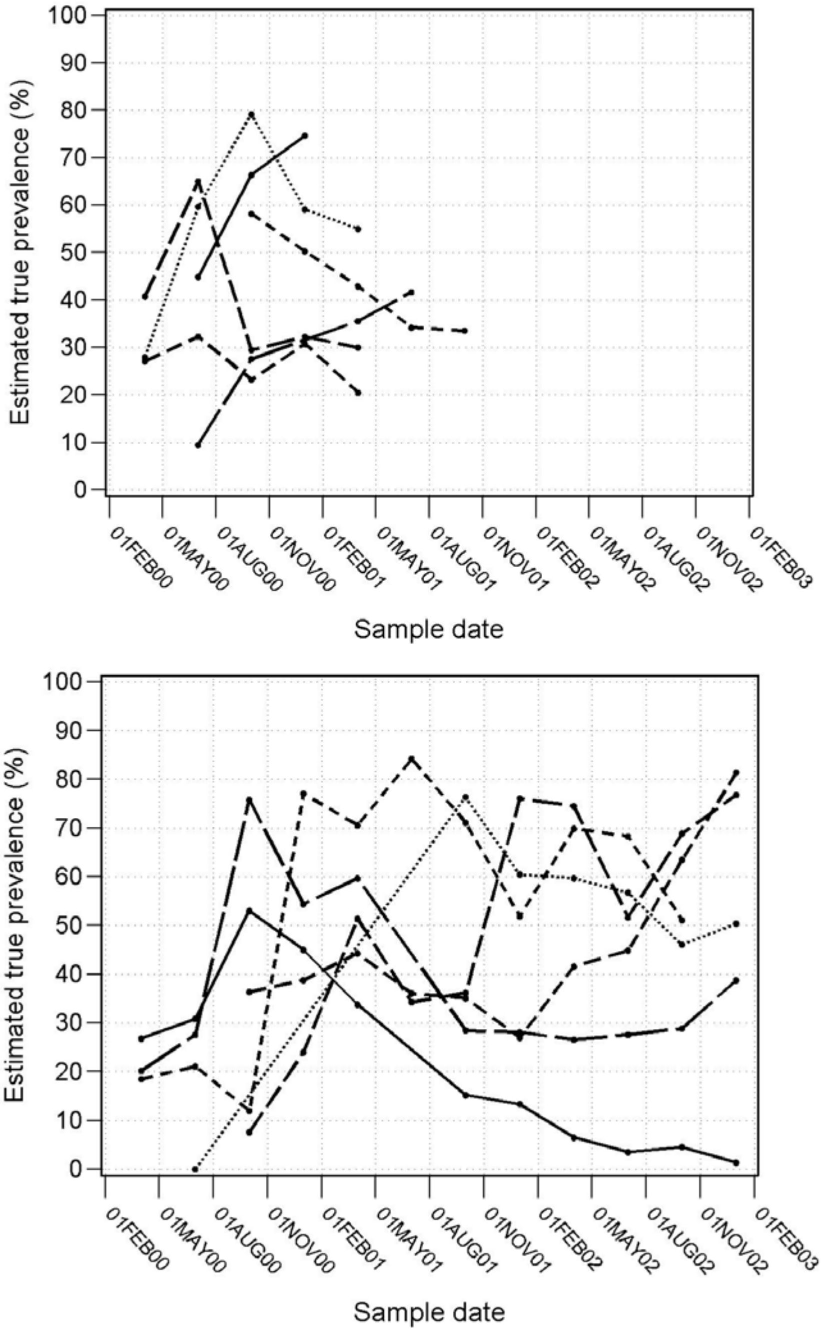


Fig. 3 True prevalence estimates of *Salmonella* Dublin infection over a 1-3 year study period in 12 infected Danish dairy herds. The 6 herds in the top graph were sampled for a period of 1-½ year. The 6 herds in the bottom graph were sampled for 2-3 years in total.

Table 2. The resulting model using logistic regression analysis of risk factors for becoming a carrier as opposed to becoming transiently infected upon infection with *Salmonella* Dublin in 265 animals from 12 infected Danish dairy herds in the years 1999-2003.

	ESTIMATES				
RISK FACTORS	β	S.E.	P[a]	Odds ratio (OR)	95% CI of OR
Intercept	-0.281	0.85			
Age and time from calving group			0.021		
Calves 0-1 year old	0.602	0.70		1.8	0.5-3.2
Heifers 1 year to 70 days before 1st calving	3.216	1.12		24.9	22.7-27.1
Heifers and cows ±70 days from 1st calving	0.999	0.57		2.7	1.6-3.8
1st parity cows in mid to late lactation	1.089	0.47		3.0	2.1-3.9
Cows 2nd or higher parity ±70 days from calving	-0.478	0.79		0.6	0-2.2
Cows 2nd or higher parity in mid to late lactation	0			1.0	
Herd prevalence (TP[b])	-0.031	0.01	0.009	0.73[c]	0.7-0.8
Random effect	Variance component	S.E.	P[d]		
Herd	4.61	2.67	0.042		

[a] P-value for each factor in the overall Type 3 statistic tests
[b] TP is the estimated true prevalence of *Salmonella* Dublin infected animals in the whole herd
[c] Odds ratio per 10% increase in herd prevalence
[d] Significance level of the variance component, herd, according to the Wald test statistics

DISCUSSION

In the present study, a case-control design have been used to make inference about risk factors. However, far from all animals from the 12 herds could be included in the study due the selection criteria. Misclassification leading to biased results might be an issue. Therefore, model validation on another dataset is needed. The model has been tested with different datasets with slightly changed criteria including a few other herds and some other animals, but the main part of the data are overlapping. Therefore, a full model validation has not yet been performed. The alternative dataset lead to slightly changed model parameter estimates, but the direction and overall magnitude of the model parameter estimates remain the same supporting the conclusions (Nielsen et al., 2003b). In the present study no effect of season was seen. However, another study has indicated that season may be of importance, so this risk factor should be tested in the model, preferably in studies covering several years of repeated sampling in several herds.

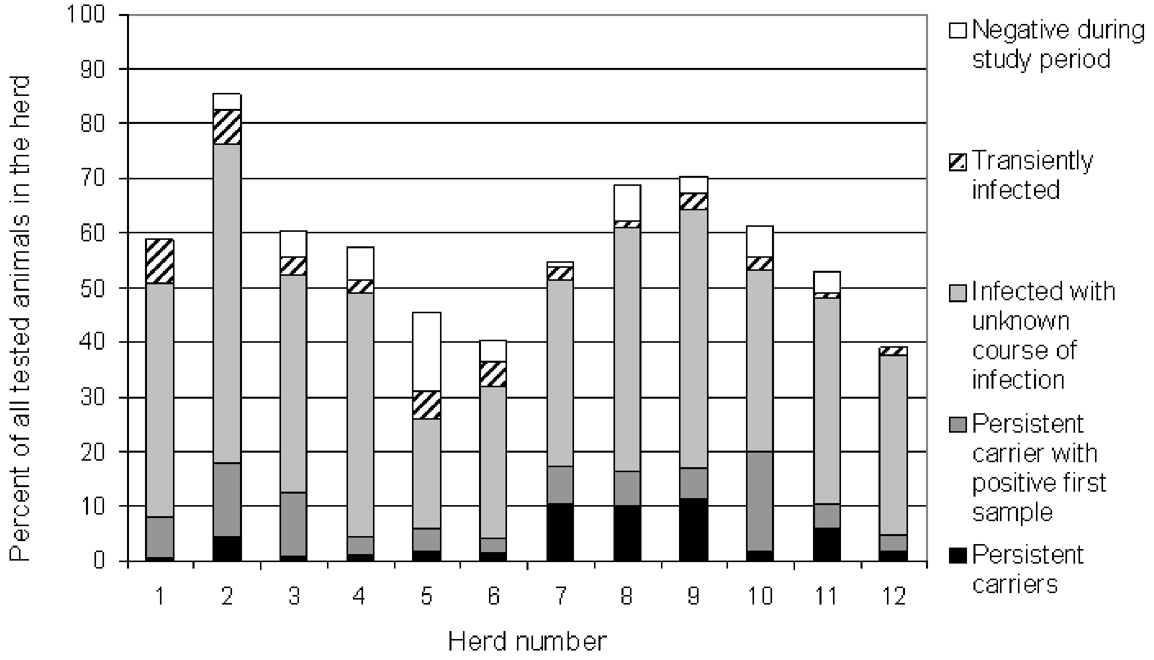


Fig. 4 Distribution of animals in different infection groups according to laboratory test results. Animals with too few samples to categorise are not shown in this graph.

Some studies indicate that though long term carriers of *Salmonella* Dublin most commonly have long term antibody responses, a few may have a very vague immunological response. For this reason risk of misclassification between carrier animals and transiently infected is possible in the present study (House et al., 1993; Hoorfar et al., 1996). Testing animals more frequently and over a longer period of time can reduce this misclassification risk (i.e., sampling every month instead of every 3 months over more than one year). It is highly recommended that no more than 1-2 months are allowed between sampling of every animal, because it is possible to miss transient infections in animals that clear the infection and have very short measurable antibody responses. Also, it is possible that some of those animals that are categorised as persistent carriers with the criteria and sampling scheme used in the present study are in fact animals that clear the infection and become re-infected at least once, and thus produce antibodies against new infections repeatedly. The high variability in the individual antibody responses makes it difficult to avoid this situation and those misclassifications that follow, but more frequent sampling is one way to minimise this problem. Other types of diagnostic tests based on measuring the avidity (the strength the binding of antibody to the agent) may aid in the distinction between true persistent carriers and re-infections.

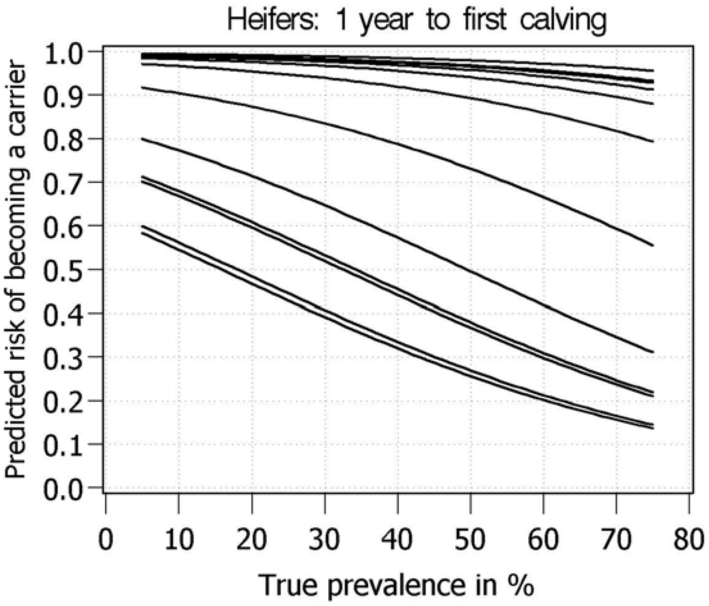


Fig. 5 Predicted risk of becoming a persistent carrier upon infection with *Salmonella* Dublin according to a model including herd prevalence at time of infection, age and time from calving of the individual animal at time of infection as fixed effects and herd as a random effect. This figure illustrates the predicted risk for calves below the age of 1 year in the 12 study herds in top graph and for heifers from 1 year of age to 70 days before first calving in the bottom graph.

The results of this study showed that cattle that become infected as heifers, around first calving or during 1st parity have significantly higher odds of becoming carriers than do cows in 2nd or higher parity and young calves. In particular, heifers between 1 year of age and up to 70 days before first calving appear to have a very high risk of becoming a persistent carrier upon infection. It is possible that this is due to hormonal changes and stress in relation to the first pregnancy and first calving that impair the ability of the heifers' immune system to combat the infection. This theory is supported by another study in which dexamethasone injections in experimentally infected animals were used to induce long term mammary gland carriers of *Salmonella* Dublin (Spier et al., 1991).

A low estimated herd prevalence of *Salmonella* Dublin was found to be associated with a higher risk of carrier production than a high herd prevalence (see Fig. 5). Care must be taken for the interpretation of this result. It does not mean that more animals become infected with *Salmonella* Dublin when the herd prevalence is low, but it means that those that do become infected when the herd prevalence is low have a significantly higher risk of becoming persistent carriers. The reasons for this phenomenon is not known, but it is supported by another study (Steinbach et al., 1996). It could be speculated that a certain threshold of infection dose is required for a sufficient cell-mediated immune response to eliminate the organism. If such a threshold (which may vary between animals due to differences in acquired immunity and genetic resistance) is not reached, lack of immediate host response would allow the organism to access the lymphoid tissues and establish longterm, protected intracellular proliferation in macrophages. It could also be speculated that those animals that are unable to resist the infection even when the infection load in the herd is low have an general impaired immunity leading to no clearance of the infection.

Finally, a significant variation between herds in the overall risk of carrier production was found in the present study. This may be due to differences in management, feeding strategies and genetic composition in the herds that lead to differences in infection load, stress load and genetic resistance.

ACKNOWLEDGEMENTS

The authors wish to thank the farmers, veterinarians and technicians for great coorporation throughout the sampling period. “The Kongeå-project” and the Danish Dairy Board provided funding for the project. We also acknowledge Ynte H. Schukken and Yrjo T. Grohn, Cornell University, for collaboration on the initial modelling in Fall 2002.

REFERENCES

Andersen, H.J., Aagaard, K., Skjøth, F., Rattenborg, E. and Enevoldsen, C. (2000). Integration of research, development, health promotion, and milk quality assurance in the Danish Dairy Industry. *In*: Salman, M.D., Morley, P.S., Ruch-Galie, R. (Eds.), Proceedings of the 9th Symposium of the International Society of Veterinary Epidemiology and Economics, pp. 258-260

Collett, D. (1991). Model checking. *In*: Modelling Binary Data, 1st Ed. Chapman & Hall, London

Fierer, J. (1983). Invasive *Salmonella* dublin Infections Associated With Drinking Raw Milk. West. J. Med. 138, 665-669

Helms, M., Vastrup, P., Gerner-Smidt, P. and Mølbak, K. (2003). Short and long term mortality associated with foodborne bacterial gastrointestinal infections: registry based study. Br. Med. J. 326, 357-361

Hoorfar, J., Feld, N.C., Schirmer, A.L., Bitsch, V. and Lind, P. (1994). Serodiagnosis of *Salmonella* dublin infection in Danish dairy herds using O-antigen based enzyme-linked immunosorbent assay. (Published erratum appears in Can. J. Vet. Res. 1995, 59 p. 25). Can. J. Vet. Res. 58, 268-274

Hoorfar, J., Wedderkopp, A. and Lind, P. (1996). Comparison between persisting anti-lipopolysaccharide antibodies and culture at postmortem in salmonella-infected cattle herds. Vet. Microbiol. 50, 81-94

House, J.K., Smith, B.P., Dilling, G.W., Da Roden, L., Spier, S.J. and Picanso, J. (1991). Serologic detection of *Salmonella* dublin infection in cattle. *In*: Proceedings of the Ninth Annual Veterinary Medical Forum., pp. 563-566

House, J.K., Smith, B.P., Dilling, G.W. and Roden, L.D. (1993). Enzyme-linked immunosorbent assay for serologic detection of *Salmonella dublin* carriers on a large dairy. Am. J. Vet. Res. 54, 1391-1399

Martin, S.W., Meek, A.H. and Willeberg, P. (1987). Veterinary Epidemiology, 1st Ed. Iowa State University Press, Ames, Iowa

Nielsen, L.R. (2003). *Salmonella* Dublin in dairy cattle: Use of diagnostic tests for investigation of risk factors and infection dynamics. Ph.D. Thesis. The Royal Veterinary and Agricultural University, Copenhagen, Denmark

Nielsen, L.R., Rattenborg, E. and Nielsen, J. (2003a). National surveillance program for *Salmonella* Dublin in Danish cattle. *In*: Proceedings of the 10th Symposium of the International Society for Veterinary Epidemiology and Economics, Viña del Mar, Chile. Abstract number 847

Nielsen, L.R., Schukken, Y.H., Grohn, Y.T. and Ersbøll, A.K. (2003b). *Salmonella* Dublin infection in dairy cattle: Risk factors for becoming a carrier. Submitted

Richardson, A. (1973). Transmission of *Salmonella* Dublin to Calves from Adult Carrier Cows. Vet. Rec. 92, 112-115

Rings, D.M. (1985). Salmonellosis in Calves. Vet Clin. North Am. Food Anim. Pract. 1, 529-539

Spier, S.J., Smith, B.P., Cullor, J.S., Olander, H.J., Da Roden, L. and Dilling, G.W. (1991). Persistent Experimental *Salmonella* dublin Intramammary Infection in Dairy Cows. J. Vet. Int. Med. 5, 341-350

Steinbach, G., Koch, H., Meyer, H. and Klaus, C. (1996). Influence of prior infection on the dynamics of bacterial counts in calves experimentally infected with *Salmonella* dublin. Vet. Microbiol. 48, 199-206

Wray, C. and Davies, R.H. (2000). Salmonella infections in cattle. *In*: Wray, C., Wray, A. (Eds.), Salmonella in Domestic Animals, 1st Ed. CABI Publishing, New York, New York State, pp. 169-190

Wray, C. and Snoyenbos, G.H. (1985). *Salmonella* dublin infection of cattle in England and Wales: its epidemiology and control. *In*: Snoyenbos, G.H. (Ed.). Proceedings of the International Symposium on Salmonella, New Orleans, pp. 173-181

ANTIMICROBIALS

QUANTITATIVE ANALYSIS OF ANTIBIOTIC CONSUMPTION IN TURKEY BROILER PRODUCTION

C. CHAUVIN* AND F. MADEC

SUMMARY

A study was carried out during 2001 on 130 turkey broiler farms to quantify and analyse antibiotic consumption during the previous nine months. A questionnaire dealing with farm, flock characteristics and health management was completed by in-person interview. Antibiotic purchases were recorded through an inventory of invoices. Quantities were expressed in three ways: total cost in euros per square metre, amounts of active substance used in grams/m² and by the number of daily-doses/m². Initially, amounts were expressed in the three different units and compared. The quantity of antibiotic used was then introduced as a variable in a linear regression analysis that considered data collected in the questionnaire as potential explanatory variables.

The quantities of antibiotic used per flock expressed in the three units were correlated but with a high heteroscedasticity. The linear mixed model fitted to the square root of the number of daily-doses/m² included five significant ($P<0.05$) explanatory variables. Variables that lower the level of antibiotic usage were thorough cleaning of the floor of the facility between flocks, changing clothes at the entrance of the facility, and administering bacterial flora. Greater antibiotic use was associated with routine prophylactic treatment, and with decreasing levels of technical and veterinary involvement in the decision to medicate flocks.

INTRODUCTION

The benefit of surveillance of antibiotic consumption in animal production has been emphasized recently (Nicholls et al., 2001). The data needed for 'antibiotic resistance risk analysis' are primarily ecological, describing antibiotics usage in animal production at a national level. Nevertheless, more detailed data are also required, particularly to identify the pattern of antibiotic use on farms. Understanding the variability of antibiotic use on farms, and the causes of this variation, would be helpful. Once risk factors are identified, interventions may be planned to influence consumption, and so reduce unnecessary use. Previous studies of antibiotic consumption in animal production showed an important variation in amounts used on farms (Nielsen et al., 2002) but no study included turkey broilers, although economic studies of turkey broiler production reported a great variation in health expenditure between farms (Anon., 2000a). An important part of this expenditure relates to antibiotics (Olivier et al., 2002), which suggests a great variation in antibiotic costs in turkey broiler production.

---

* Claire Chauvin, French Agency for Food Safety AFSSA, Zoopôles les Croix, B.P. 53, 22440 Ploufragan, France. Email: c.chauvin@ploufragan.afssa.fr

In order to measure antibiotic consumption on farms, a reliable unit of measurement must be defined. Several units can be used (Chauvin et al., 2001) such as an economic unit expressing the cost of antibiotics, a weight unit expressing the amount of active substance acquired to treat the animals, or a unit expressing the number of daily treatment doses used. Costs are easy to collect from invoices, whereas commercial names, quantities used and compositions are needed to calculate the weight of active substance consumed and number of daily-doses administered. However, it is unclear if all these units can provide the same information.

An on-farm study was carried out to assess and compare the variation in antibiotic use across turkey broiler production units, and to explore putative factors which could explain the variation in antibiotic utilisation.

MATERIALS AND METHODS

Study sample

A random selection of 130 farms in the departmental farming registers from three French regions (Centre, Bretagne, Pays de la Loire) with a reserve list of 10 farms, was completed. These regions represent 46.2%, 20.7% and 7.1% respectively of the national turkey production (CIDEF, 2002), and were conveniently located near the enquiry centres. The selection was made using these regional proportions, and represented about 5% of all specialised turkey farms. Rare products such as 'light turkeys' were excluded. The farmers were telephoned to explain the purpose of the enquiry and to obtain their agreement to participate in the study. Two farmers refused and were replaced from the reserve list.

Data collection

During the first telephone call, an appointment was made to visit the farm between March and July 2001 so as to enable data collection using face-to-face interviews and to allow a study of invoices. Both tasks were performed by ten experienced interviewers who had been previously trained prior to the commencement of the study. The interviewers were affiliated to regional professional agricultural bodies. During each farm visit, the interviewers initially completed a structured questionnaire (Table 1), which had been used routinely by investigators for several years (Anon., 2000a) to collect information about farm characteristics, farmer's attitude and habits towards health management, and on the flocks' technical performances. This questionnaire was pilot-tested by interviewers on a separate random sample of five flocks, and this led to only small modifications. Antibiotic use was also investigated through an inventory of medication purchases. For each farm, invoices were consulted and commercial names, presentation and quantities were recorded for the turkey flocks reared and slaughtered during the last 9 months (between 1/7/2000 and 31/3/2001). On farms with more than one flock, purchases were attributed to a flock according to the farmer's recollection and supporting evidence such as date of invoicing. For each flock, the study time frame for invoices was extended to 15 days prior to the flock's arrival in order to include any medicines purchased in advance of the arrival of the birds.

Table 1. Summary of items included in the questionnaire.

Items	Data collected
General questionnaire	
Farmer	Identity, age, experience, education
Farm	Land area for crops, other animal production
Turkey broiler production	Characteristics of facility (number of birds, surface area, equipment…)
Flock	Production type, technical characteristics (age at slaughter, density, …), feed type, sanitary events and mortality, economic results
Health management practices	
Medication purchase	Sellers characteristics, health expenditures
Prophylactic treatments	Nature, age at treatment, reasons…
Treatment management	Treatment decision process, administration…
Biosecurity practices	Cleaning and disinfection procedure, hygiene implementation

Variable definition

The total amounts of antibiotic bought for each flock were expressed in three different units:

1. Expenses were summed for each flock and converted from French francs into Euros.

2. Using the quantities and commercial names recorded on the invoices along with the corresponding composition (Anon., 2000b), purchased medications were converted into grams (g) of active substance. When necessary, different chemical forms of an active substance were converted into a reference activity using standard conversion rules.

3. To more accurately define the therapeutic use of medications, the amounts of active substance were converted into the number of turkey 'defined daily doses' (DDD) used (Chauvin et al., 2001). Amounts of each antibiotic type were divided by the daily dosage required by an average turkey broiler. This dosage was calculated according to the recommended dosage (Anon., 2000b) for a broiler weighing 3kg, which was found to be the mean weight at treatment of turkey broilers in a large sample collected earlier (Chauvin et al., 2002).

To compare antibiotic consumption between flocks, the quantities expressed in these three units (€, g, DDD) were quasi-standardized by dividing them by the facility's surface area expressed in square metres, which is a widely used denominator in technical and economic analysis of broiler poultry production (Anon., 2000a). Lastly, these values were normalized using an appropriate transformation, whose validity was checked by means of the Shapiro-Wilk and Kolmogorov-Smirnov tests performed with the SAS software (SAS Institute Inc., 2001).

Statistical analysis

The relationship between the three units used was explored graphically by contour-plot drawing and by correlation coefficient calculation. One unit was chosen according to observations made at this stage. Rearing practices or farm characteristics, which were significantly related to antibiotic consumption, were identified using a linear mixed regression analysis performed in two stages. In the first step, variables were screened individually, as were all interactions between them. For pairs of variables showing strong structural colinearity, the one more closely related to the amount of antibiotic used was chosen. Variables which passed this first stage ($P<0.25$) were considered in a multivariate linear model. A step-by-step backward selection procedure was applied ($P<0.05$) to obtain a final model. To account for the farm cluster effect (due to the fact that several flocks from the same farm could be incorporated into the study), a 'farm' random effect was introduced in the model. Calculations were performed using the MIXED procedure of the SAS system (SAS Institute Inc., 2001).

RESULTS

Data sample

Within the study, 246 turkey broiler flocks were included representing 3.2% of the flocks reared in France during the same period (CIDEF, 2002). An average of 1.9 flocks was studied per farm (range 1-3, standard error = 0.4). Questionnaires and invoice details were available for all of them.

Comparison of the units

For all three units (€/m^2, g/m^2 and DDD/m^2), the distribution of the amounts of antibiotic used was characterised by many flocks with no or low use, and the number of flocks decreased rapidly as the amounts increased. The square root transformation allowed normalisation of the distributions according to the Shapiro-Wilk and Kolmogorov-Smirnov tests. Figure 1 represents the contour-plot of the antibiotic usage of the 246 flocks expressed in the three units. Pearson correlation coefficients and their associated P value are given in Table 2.

Table 2. Pearson coefficients (lower left) and associated P-value (upper right) for antibiotic consumption expressed in three different units.

	$\sqrt{(€/m^2)}$	$\sqrt{(g/m^2)}$	$\sqrt{(DDD/m^2)}$
$\sqrt{(€/m^2)}$	-	<0.0001	<0.0001
$\sqrt{(g/m^2)}$	0.67	-	<0.0001
$\sqrt{(DDD/m^2)}$	0.65	0.86	-

Fig. 1 Contour plot of the antibiotic consumption of 246 turkey broiler flocks expressed in three different units: X axis corresponds to the square root of the number of DDD/m², Y axis corresponds to the square root of the total cost in €/m², Z axis corresponds to the square root of the weight of active substance in g/m². (Coloured lines represent the Z dimension, increasing from violet to red, with a same colour indicating the same Z value. Black plots represent the flocks).

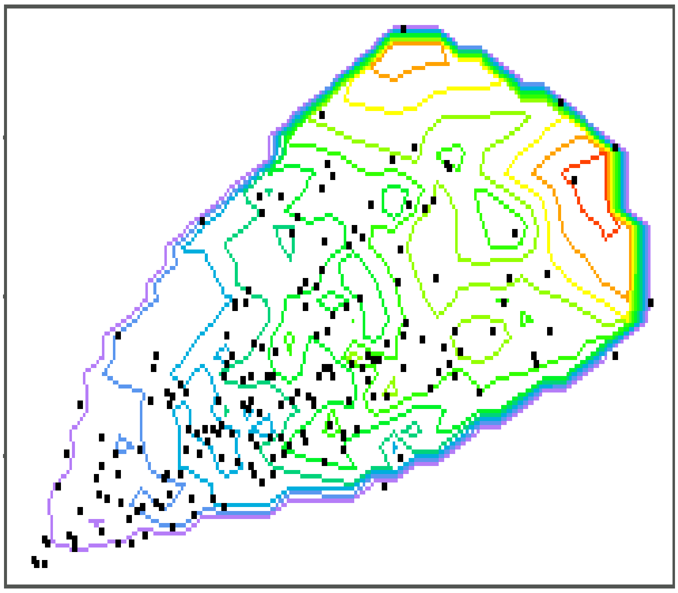


Table 3. Final linear mixed model of antibiotic use in turkey broiler flocks.

Variable	Estimate	SE	P
Intercept	7.8	0.77	<0.0001
Floor of the facility thoroughly cleaned between flocks			
yes	- 2.56	1.25	0.04
no	0	-	-
Clothes changed at the entrance of the facilities			
yes	-1.59	0.72	0.03
no	0	-	-
Systematic prophylactic antibiotic treatment			
yes	2.54	0.85	0.004
no	0	-	-
Systematic bacterial flora administration			
yes	-4.76	1.28	0.0003
no	0	-	-
Antibiotic treatment requested by			
Farmer	2.45	1.13	0.03
Technician	1.45	0.76	0.03
Veterinarian	0	-	-

Antibiotic consumption analysis

The unit selected for the linear regression analysis was the square root of the number of DDD per square metre. The final linear mixed model obtained is given in Table 3. Model residues were normally distributed and residual variance remained important (variance of the farm random effect = 9.12; residual variance = 11.46). Five parameters all related to hygienic practices and health management were retained in the final model. In all cases, practices ensuring better hygiene were associated with lower antibiotic use.

DISCUSSION

To measure antibiotic consumption in the studied broiler flocks, three units were used providing different types of information (Chauvin et al., 2001). Even if the units were found to be correlated, an important heteroscedasticity was noticeable on the contour-plot, and so the units could not be considered as equivalent. In our opinion, in order to investigate differences in antibiotic use between farms or flocks, a unit reflecting the therapeutic use of antibiotics and the 'treatment pressure', such as the DDD, would be preferable. This unit was chosen for the second part of the present study. In an economic context, in order to explore ways to reduce expenditure on antibiotics, costs should be used, and a second analysis should be performed.

Some farm practices were found to be linked to the amounts of antibiotics used. Few studies have been carried out on this topic (Nielsen et al., 2002), and few risk factors were identified in this study. Variables retained in the final model were mainly related to good hygienic practices, such as changing clothes at the entrance of the facility and thorough cleaning of the floor. These factors can be considered as markers of good farming practice, aimed at preventing disease introduction and so reducing drug use. Other variables were related to medication management and use. Systematic bacterial flora administration was found to be associated with lower use of antibiotics. Bacterial flora are administered mainly at periods of risk for digestive disorders such as after vaccinations or during transition between different feeds. They are known to help prevent digestive disorders (Patterson & Burkholder, 2003). On the other hand, routine prophylactic use of antibiotics rather than therapeutic use was finally found to be associated with greater use of antibiotics. Prophylactic use of antibiotics was considered to be routine when it did not follow observation of clinical symptoms or an increase in mortality, and only when it was repeated on several successive flocks. The main reason reported for such treatment was a repeated experience of disease in previous flocks. However, the effectiveness and timing of treatment never appears to have been assessed or questioned. The farmers' attitudes towards antibiotic use could be assessed further by whether the decision to treat was made by the farmer, technician or veterinarian. Surprisingly, this variable remained in the model. This parallels a previous observation made in human medicine where physicians were found to increase the rate of prescription of antibiotics for children's infections at the insistence of the parents (Mangione-Smith et al., 1999).

However, it must be highlighted that the linear model did not explain the major part of the variation. Therefore, several other factors may influence antibiotic use which were not taken into account within this study. Farmers need to be relayed information about factors affecting antibiotic use and misuse, as identified in this study, and particularly about the value of good hygiene practices.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the farmers and the investigators for their active co-operation in this study.

REFERENCES

Anon. (2000a). Résultats de l'enquête 1999-2000. Editée par la Chambre d'agriculture du Morbihan avec la participation des Chambres d'Agriculture du Grand Ouest. 43pp.

Anon. (2000b). Dictionnaire des medicaments veterinaries et des produits de santé animale. 10ème édition, Veillet F., Vandaële E., (Eds). Editions du Point vétérinaire, 1814pp. Maisons-Alfort, France.

Chauvin, C., Hardy, A., Le Bouquin, S. and Orand, J.P. (2002). Première base de données vétérinaires française – le projet O.A.C.A. en filière volailles. 2nd colloque de pharmaco-épidémiologie, 22-23 octobre 2002, Paris

Chauvin, C., Madec, F., Guillemot, D. and Sanders, P. (2001). The crucial question of standardisation when measuring drug consumption. Vet. Res. 32, 533-543

CIDEF (2002). La Dinde par les chiffres – 2002, 42pp.

Mangione-Smith, R., McGlynn, E.A., Elliott, M.N., Krogstad, P. and Brook, R.H. (1999). The relationship between perceived parental expectations and pediatrician antimicrobial prescribing behaviour, Pediatrics 103, 711-718

Nicholls, T., Acar, J., Anthony, A., Franklin, R., Gupta, Y., Tamura, Y., Thompson, S.R., Threlfall, J.E., van Vuuren, M., White, D., Wegener, H. and Costarrica, L. (2001). Antimicrobial resistance: monitoring the quantities of antimicrobials used in animal husbandry. Rev. Sci. Tech. 20, 841-847

Nielsen, J.P., Petersen, H.H. and Bak, H. (2002). Antimicrobial consumption in Danish finishing herds, 17th IPVS congress, Iowa, Ames, USA.

Olivier, B., Bouvarel, I. and Berthelot, A. (2002). Etat des lieux des pratiques vétérinaires en élevage de dindes de chair. Sciences et Techniques avicoles 40, 21-28

Patterson, J.A. and Burkholder, K.M. (2003). Application of prebiotics and probiotics in poultry production. Poult. Sci. 82, 627-631

SAS Institute Inc. (2001). SAS/STAT User's Guide, version 8. SAS Institute, Cary, NC.

SPATIAL POINT PATTERN ANALYSIS TO STUDY THE RESISTANCE BEHAVIOUR OF *ENTEROCOCCUS* TO TETRACYCLINE IN BULK MILK

K. FUCHS*, A. DEUTZ, P. WAGNER AND J. KÖFER

SUMMARY

In 2002 and 2003, 752 samples of bulk milk were tested for their resistance behaviour of indicator bacteria (*Enterococcus*, *E. coli*) to several antibiotics. A spatial point pattern may be thought of as a consisting of a set of locations (s_1, s_2, …) in a defined 'study region', R, at which 'events' of interest have been recorded. In this paper events are of two different types (bivariate point pattern): a set of events which show resistance behaviour and those without. To exemplify the use spatial point pattern analysis, the resistance behaviour of *Enterococcus* to tetracycline is studied. Kernel density estimation and K functions are used to investigate whether the distribution of the resistant samples clusters relative to the sensitive ones and to locate high and low density areas for resistance behaviour of *Enterococcus* to tetracycline.

INTRODUCTION

The development of antibiotic-resistant strains of bacteria in livestock is not only a major concern of veterinary medicine but is also considered as a source of resistant pathogens in human medicine. According to "Good Veterinary Practice (GVP)" the responsibility of veterinarians for consumer health is defined as ranging from diagnosis, identification of indications, drug selection, compliance with dosage regulations and documentation of drug use to the monitoring of therapeutic success.

The EU Council Resolution of 8th June 1999 on antibiotic resistance entitled "A strategy against the microbial threat" calls on Member States to develop multidisciplinary and cross-sectoral strategies to control antibiotic resistance. The emergence of antibiotic resistance in animal pathogens is closely connected with therapeutic, prophylactic and metaphylactic measures in animal husbandry. The antibiotic resistance of zoonotic pathogens, such as *Salmonella*, *Campylobacter* spp., *Listeria* or pathogenic *E. coli* in food of animal origin is also of increasing significance. The determination of the resistance behaviour of so-called indicator bacteria, i.e., commensals such as *Enterococcus* or *E. coli* omnipresent in the intestinal flora, is also a priority because they are considered as a reservoir of resistant strains in the intestines and are capable of transferring resistance genes to both animal and zoonotic pathogens. The transmission of resistant bacteria from animals to humans may occur through food consumption or direct contact. These bacteria are capable of transferring their resistance determinants (e.g. plasmids or transposons) to the naturally occurring bacteria of the human intestinal flora.

---

* Klemens Fuchs, Institute for Applied Statistics, Joanneum Research, Steyrergasse 25a, 8010 Graz Austria. Email: klemens.fuchs@joanneum.at

In view of the topicality of the issue, the Department of Veterinary Administration of the Styrian Government established a resistance monitoring programme in 1999 after extensive preparatory work in order to ensure effective consumer protection (Köfer et al., 2002). It is part of an integrated control system for the improvement of food safety and includes both livestock and the meat produced from it.

To explore possible spatial patterns within Styria spatial point pattern algorithms were applied to analyse the resistance data.

MATERIALS AND METHODS

Sample size

The sampling system in the bulk milk study was based on a representative sampling model, which involved the taking of random samples (every eighth Styrian cattle breeding farm) within the scope of the BVD control programme (bulk milk analyses). All results of the resistance analysis from bulk milk samples were assigned to the respective farms by means of their LFBIS (information system for agricultural and forestry enterprises) numbers and evaluated using a geographical information system (VETGIS$^{©}$-Styria).

Detection methods

Tests and analyses were performed on bulk milk. For this purpose 2 ml milk were taken within the BVD control programme (bulk milk analyses). The samples were cooled and subsequently conditioned.

The CATC medium (Merck 1.10279) served as selective medium for the determination of *Enterococcus*. Red *Enterococcus* colonies developed in the culture medium after an incubation period of 48 hours at 37°C.

Resistance test – determination of MIC values

The resistance test and determination of MIC (minimum inhibitation concentration) values were done using SenistitreR (MCS-Diagnostics, Veldpoort 28, NL-6071 JL Swalmen), a commercially available MIC technique using dehydrated antimicrobials in micro titre wells. The wells were inoculated according to NCCLS guidelines and incubated aerobically at 37°C for 18-22 hours. The MIC was defined as the lowest concentration of antimicrobial with no visible growth. A sample was classified as resistant, if the MIC of the isolate exceed the appropriate breakpoint (Aarestrup et al., 1998). The breakpoint for tetracycline in bulk milk, for example, was determined at 16.

Statistical background

A spatial point pattern may be thought of as a consisting of a set of locations (s_1, s_2, …) in a defined 'study Region', R, at which 'events' of interest have been recorded. In the simplest case the data set comprises solely the event locations. In this study, events were of two different types (bivariate point pattern): a set of events which showed resistance behaviour and those which did not. The simplest theoretical model for spatial point pattern is that of complete spatial randomness (i.e., homogeneous Poisson process) in which the events are distributed independently according to a uniform probability distribution over the region, R (Diggle, 1983).

Looking at the data-set, it made little sense to compare observed spatial clusters of resistance behaviour against an homogeneous Poisson model. We investigated whether the distribution of the resistant samples clustered relative to the sensitive ones and attempted to locate high and low density areas for resistance behaviour of *Enterococcus* to tetracycline.

Kernel estimation

From a statistical point of view, an observed spatial point pattern can be thought of as the realisation of a spatial stochastic process. The behaviour of general spatial stochastic processes may be characterized in terms of its first- and second-order moments. First-order moments are described in terms of the intensity $\lambda(s)$ of the process, which is the mean number of events per unit area at the point **s** (Diggle, 1983). Several methods exist to estimate the first-order intensity (Kaluzny et al., 1997). One of the most popular is the kernel-density estimation.

An exploratory tool for examining the first-order properties of a point process could be a count of events per unit area within a moving 'window'. A window, defined as being of fixed size is centred on a number of locations in turn, where these are arranged in a fine grid superimposed over R. We thus obtain estimates of the intensity at each grid point. Kernel estimation is a generalization of this idea, where the window is replaced with a moving three-dimensional function (kernel) which weights events within its sphere of influence according to their distance from the point at with the intensity is being estimated. As to the exact functional form of the kernel, k(), we require a decreasing radially symmetric bivariate function providing a total weight of unity over the region of influence. A typical choice might be the so-called quadratic kernel. Then the estimate of $\lambda_\tau(\mathbf{s})$ may be simply expressed as

$$\widehat{\lambda}_\tau(s) = \sum_{d_i<\tau} \frac{3}{\pi\tau^2}(1-\frac{d_i^2}{\tau^2})^2 \qquad (1)$$

where d_i denotes the distance between event and point s and τ is the bandwidth (f.e. radius).

Notice that the kernel estimation is very sensitive to the choice of τ and that the so-called "edge effects" tend to distort the kernel estimates. To overcome these problems we used adaptive (i.e., local adjustment of τ) edge-corrected kernels.

The K function

If a point process is stationary (and isotropic) there is a close mathematical relationship between the second-order intensity and an alternative characterization of second-order properties known as the K function. This is defined by the relationship $\lambda K(d) = E$ (#(events $\leq$ distance d of an arbitrary event)), where K(d) describes the extent of which there is spatial dependence in the arrangement of events.

We have already made reference to the idea of a random arrangement of events. Formally, the point process that gives rise to such an arrangement is called a homogeneous Poisson process. We say that an arrangement of events shows complete spatial randomness if it is a realization of such a process. As far as the K function for a complete spatial random process is concerned, the important point is that the probability of the occurrence of an event at any point in R is independent of what other events have occurred and is equally likely over the whole area of R. For a homogeneous process with no spatial dependence, the expected number of events with the distance d of a randomly chosen event is simply $\lambda\pi d^2$. In other words $K(d) = \pi d^2$, representing a circle with radius d. If there is clustering of point events, we would expect to see an excess of events at short distances. For small values of d, the observed value of K(d) will be greater than πd^2.

A suitable edge-corrected estimation of the K function is given by (Boots & Getis, 1988)

$$\hat{K}(d) = \frac{1}{\lambda^2 R} \sum\sum_{i \neq j} \frac{I_d(d_{ij})}{w_{ij}} \tag{2}$$

where R is the area of region R, $I_d(d_{ij})$ is an indicator function that takes the value 1 when d_{ij} is less than d, and w_{ij} is the proportion of the circumference of this circle which lies within R.

Once calculated, $\hat{K}(d)$ can be compared with its expected form according to particular theoretical situations.

Extensions to the K function: Case-Control Patterns

Given n_1 events of primary concern (cases, i.e., resistant) and n_2 events representing the "natural" homogeneity (controls, i.e., sensitive) then, in the absence of clustering among the cases relative to the controls, if we pool the two sets of events we would expect the n_1 case 'labels' to be attached at random to the combined set of events; this is called a random labelling of events (Gatrell et al., 1996). Under such random labelling it can be shown that the K functions for the cases $K_1(d)$ and for the controls $K_2(d)$ are identical.

To test the hypothesis of "random labelling", which suggests that $K_1(d) = K_2(d)$ we might use a plot of the difference $\hat{D}(d) = \hat{K}_1(d) - \hat{K}_2(d)$ against d to explore such a hypothesis. When $\hat{D}(d)$ is plotted against d, peaks in this plot will show clustering over and above that environmental heterogeneity. A more formal assessment of the significance of peaks in this plot is the idea of simulation techniques. Under the assumption of complete spatial randomness we may perform m independent simulations of n events in the study region. For each simulated point pattern we can estimate $\hat{K}(d)$ and the use the maximum and minimum of these functions for the simulated patterns to define upper and lower simulation envelope. If the estimated $\hat{K}(d)$ lies above the upper envelope, we can speak of significant spatial clustering, if it lies below the lower limit, this is evidence of regularity in the arrangements of events (Bailey & Gatrell, 1995). Such a method provides a useful complement to alternative approaches (see, for example, Cuzick & Edwards, 1990) which are essentially based on extensions to distribution function of inter-event distances.

Spatial scan statistic

Alternative tests for special clustering which do detect a range of clustering types are the space-scan statistics. The general statistical theory behind the spatial and scan statistic is described in detail by Kulldorff (1997).

The spatial scan statistic uses two different probabilistic models, based on the Bernoulli and Poisson distributions, respectively. With the Bernoulli model, there are cases and non-cases as a 0/1 variable. They may reflect cases (i.e., resistant) and controls (i.e., sensitive) from a larger population, or they may together constitute the population as a whole. With either model, the scan statistic adjusts for the uneven population density present in almost all populations, and the analysis is conditioned on the total number of cases observed. For each location and size of the scanning window, the alternative hypothesis is that there is an elevated rate within the window as compared to outside. Under the Bernoulli assumption, the likelihood function for a specific window is then proportional to

$$\left(\frac{c}{n}\right)^{c}\left(\frac{C-c}{N-n}\right)^{(C-c)}\left(1-\frac{C-c}{N-n}\right)^{(N-n)(C-c)} I() \tag{3}$$

where C is the total number of cases over the whole area, c is the number of cases within the window, n is the total number of cases and controls in the cluster and N is the total number of cases and controls in the data set. I() is an indicator function.

The likelihood function is maximized over all windows, identifying the window that constitutes the most likely cluster. This is the cluster that is least likely to have occurred by chance. The likelihood ratio for this window is noted and constitutes the maximum likelihood ratio test statistic. Its distribution under the null-hypothesis and its corresponding p-value is obtained by repeating the same analytic exercise on a large number random of replications of the data set generated under the null hypothesis, in a Monte Carlo simulation.

RESULTS

There were 752 *Enterococcus* strains isolated from these samples and tested for resistance to 13 different antibiotics, respectively. The percentage of multiresistant isolates was 15%. The proportion of resistant isolates in 2002 and 2003 were very similar. The highest rates were found for tetracycline (52%), streptomycin (21%), kanamycin (18%), flavomycin (15%) and chloramphenicol (13%).

In another survey implemented in 2002, *Enterococcus* spp. isolates from cattle faeces displayed a high level of resistance to flavomycin, whereas isolates from bulk milk samples of cows showed only a high level of resistance to tetracycline.

To exemplify the use spatial point pattern analysis we analysed the resistance behaviour of *Enterococcus* to tetracycline. Fig. 1 shows a point map of the 383 resistant (circles) and 369 sensitive (triangles) samples.

According to Eq. (1) the kernel density estimates $\lambda_1(s)$ for the resistant samples and $\lambda_2(s)$ for the sensitive ones were calculated separately. Fig. 2 shows the map of the kernel density ratio

$$\hat{p}(s) = \frac{\lambda_1(s)}{\lambda_1(s) + \lambda_2(s)}$$

The highest ratio of resistant samples can be seen in the east and some hot-spots in the north and west. To find out if these areas can be interpreted as spatial clusters K functions were estimated separately for resistant and sensitive samples (Eq. 2) and subtracted to obtain the difference function $\hat{D}(d)$. Upper and lower simulation envelopes were developed by performing 999 replications. As $\hat{D}(d)$ lies between the upper and lower envelope (Fig. 3) no statistically significant evidence for clustering can be seen

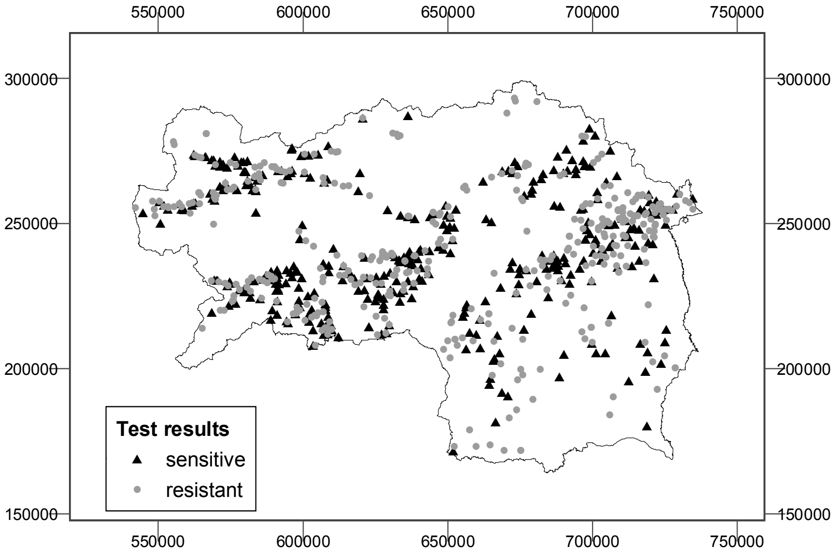


Fig. 1 Point map of resistant (circles) and sensitive (triangles) samples

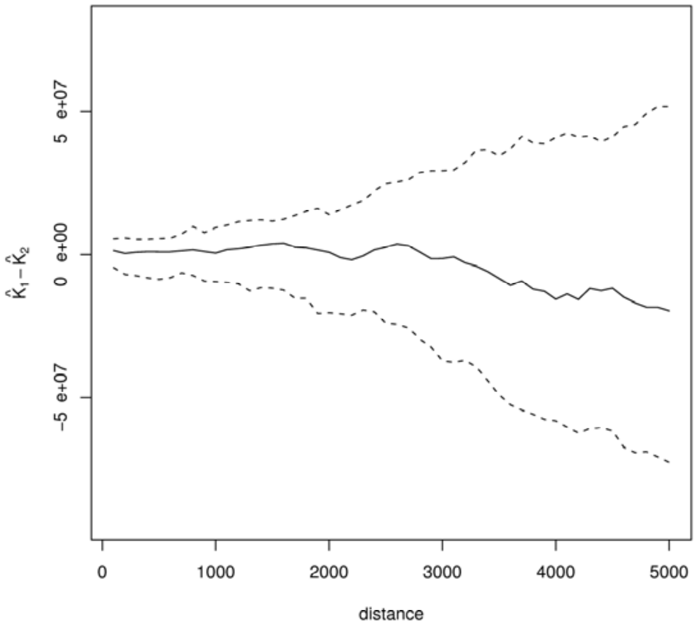


Fig. 2 Kernel density ratio for resistance behaviour

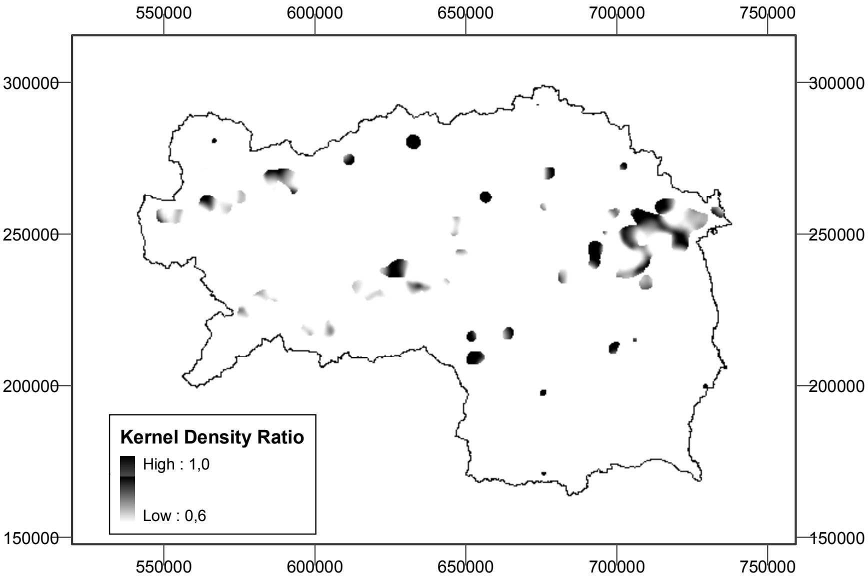


Fig. 3 Difference between K functions (solid line) and simulated envelope to test for spatial clustering

To investigate whether there were high (low) density areas of resistant samples, spatial scan statistics were calculated. Fig. 4 shows the most likely clusters for a high density area of resistant samples and a low density area, respectively. Calculations were performed using the software package SaTScanTM Ver. 3.0 (Kulldorff, 2002) with 999 Monte Carlo replications.

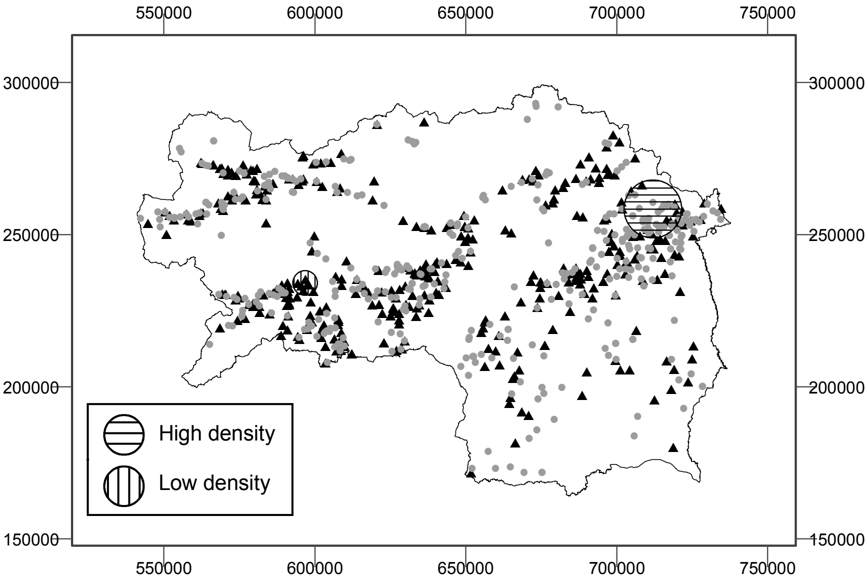


Fig. 4 High and low density area

DISCUSSION

First warnings of increasing antibiotic resistance of mastitis pathogens were voiced as early as 35 years ago. New methods of targeted treatment of mastitis infections were developed, taking into consideration predisposing environmental factors. The rapid development of antibiotic drugs was considered as a race against the increasing antibiotic resistance of infectious pathogens.

The considerable differences in the regional distribution of resistances in the Styrian area under investigation may be due to the agricultural structure (small farms, farmers working in other jobs to obtain a supplementary income in the south), wrong dosage and administration of antibiotic drugs and insufficient accompanying measures during therapy. Another reason for higher resistance rates in the east of Styria may be the combination of cattle farming and intensive pig and poultry farming in this region, assuming that bacterial strains from swine or poultry were transferred to cattle or are able to transfer resistance genes to udder pathogens, or themselves cause mastitis infection.

There is no doubt that the antibiotic treatment of mastitis infections is the main reason for the development of resistance in udder pathogens. Therefore, insufficient dosage in intra-mammary administration of antibiotics or non-indicated application of dry cow therapy are to be avoided (Deutz & Köfer, 1998). Special attention should be paid to accompanying measures, such as udder health programmes, California mastitis tests, the milking sequence and the culling of therapy-resistant infected cows. Of course, resistance in bulk milk may also result from other treatments.

The resistance testing from the bulk milk sampling represents an efficient method of the supervision of the resistance situation of indicator bacteria and zoonotic pathogens of cattle. Future risk management actions (e.g., prohibition of the use of single antibiotics) should be considered, if fixed resistance rates are exceeded.

The Styrian Resistance Monitoring Programme (REMOST) is designed to investigate the resistance behaviour of bacteria on a continuous basis. It consists of a sampling system, which indicates where, how and when samples are to be taken, an analysis system for the continuous analysis of data and a catalogue of measures based on these modules. Isolation of the bacterial strains is done by streaking the material to be tested (faeces, meat, milk) on different agar media. After biochemical verification of suspect colonies, the resistance behaviour is tested using the SENSITITRE® system of MCS-Diagnostics.

The test results are published on an annual basis and are fed into a central database, which is linked to the VETGIS© Styria geographical information system. These data are combined with current analysis results from human medicine and with international animal production data in order to assess the risk of transmission of resistance factors. Continuous data acquisition allows possible changes in the resistance behaviour of bacteria to be recognised at an early stage.

In summing up the investigations, it can be said that drug usage in the keeping of livestock must be based on the principles of Good Veterinary Practice (Van Miert, 1993). Intensified interdisciplinary cooperation between human and veterinary medicine is required to investigate the problem of resistance transfer.

REFERENCES

Aarestrup, F.M., Bager, F., Jensen, N.E., Madsen, M., Meylin, A. and Wegener, H.C. (1998). Resistance of antimicrobial agents used animal therapy in pathogenic-, zoonotic- and indicator bacteria isolated from different food animals in Denmark: A baseline study for the Danish integrated Antimicrobial Resistance Monitoring Programme (DANMAP). APMIS 106, 745-770.

Bailey, T.C. and Gatrell, A.C. (1995). Interactive spatial data analysis. Longman Higher Education, Harlow. 413p

Boots, B.N. and Getis, A. (1988). Point pattern analysis. Sage Scientific Geography Series, Vol. 8, Sage Publications, London

Cuzick, J. and Edwards, R. (1990). Spatial clustering for inhomogeneous populations. J. of the Royal Stat. Soc. Series B, 52, p. 73-104

Deutz, A. and Köfer, J. (1998). Situation and Development of Antibiotic Resistance in Swine, Cattle and Poultry. Proceedings 4. Weltkongress Lebensmittelinfektionen und -intoxikationen, 7. bis 12. Juni, Berlin, p. 157, (Proceedings p. 715-720)

Diggle, P.J. (1983). Statistical analysis for spatial point patterns. Academic Press, London. 148p

Gatrell, A.C. Bailey, T.C., Diggle, P.J. and Rowlingson, B.S. (1996). Spatial point pattern analysis and its application in geographical epidemiology. Trans. Inst. Br. Geog. 21, 256-274

Kaluzny, S.P., Vega, S.C., Cardosos, T.P. and Shelly, A.A. (1997). S+ Spatial Stats. Springer. 327p

Köfer, J., Pless, P. and Fuchs, K. (2002). Implementation of a resistance monitoring programme in Styrian meat production-Investigation of the resistance behaviour of indicator bacteria and zoonotic pathogens. Fleischwirt. Int. 2, p. 46-50.

Kulldorff, M. (1997). A spatial Scan Statistic. Commun. Statist. 26, p. 1481-1496

Kulldorff, M. (2002). SaTScan^{TM} User Guide for version 3.0. http://srab.cancer.gov/satscan/ 59p

Van Miert, A.S. (1993). Good Veterinary Practice. Proceedings of the 14th Intensivseminar d. Steirischen Schweinegesundheitsdienstes, Heiligenblut, p.154-161

ANTIBIOTIC RESISTANCE IN *CAMPYLOBACTER* SPP. IN SWISS POULTRY PRODUCTION

G. REGULA[*], U. LEDERGERBER, R. STEPHAN, J. DANUSER, D. LO FO WONG,
B. BISSIG-CHOISAT AND K.D.C. STÄRK

SUMMARY

A monitoring programme for antibiotic resistance was launched in 2002 in co-operation with the Swiss poultry industry. Resistance patterns of *Campylobacter* spp. were obtained from cloacal swab samples of live birds and neck skin samples at slaughter. The prevalence of antibiotic resistance in the *Campylobacter* strains isolated was 11% and 6% for ciprofloxacin in the years 2002 and 2003, respectively. One percent and 3% of isolates were resistant against erythromycin in both years, 4% and 4% for tetracycline, 9% and 3% for ampicillin, 2% and 4% for amoxycillin, 0.3% and 0.6% for gentamicin, 20% and 9% for sulphonamide and 18% and 14% for streptomycin.

The monitoring programme will be further developed to include other animal species and pathogens. Baseline data from the first two years of monitoring will be used to optimise the number of samples collected at different sampling points along the production process.

INTRODUCTION

In recent years, antibiotic resistance in bacteria from animal origin has been of increasing concern to consumers. The potential hazard posed to human health from zoonotic pathogens that are resistant to the antibiotics used for therapeutic treatment of human infections is of particular concern. For this reason, several countries have implemented monitoring programmes for antibiotic resistance (Wray & Gnanou, 2000). Moreover, international working groups have published recommendations for the design of antimicrobial resistance monitoring programmes for food animals (Anon., 1999; Caprioli et al., 2000). These recommendations together with the experience from existing networks form the implementation framework for the development of such monitoring programmes in countries such as Switzerland.

In order to develop efficient sampling plans, the variability of the characteristics to be measured between animals, between farms and between slaughterhouses need to be known. Dunlop et al. (1999) investigated this issue at the farm level with respect to antimicrobial resistance in finisher pigs and described a predominance of between-pig variability. By the same token, an international working group recommended selecting only one sample per farm to avoid cluster effects (Anon., 1999). This however, is only true if within-farm variability of animals is smaller than between-farm variability. The above-mentioned working group

---

[*] Gertraud Regula, Swiss Federal Veterinary Office, Schwarzenburgstr. 161, CH-3003 Bern, Switzerland. Email: Gertraud.Regula@bvet.admin.ch

acknowledged that the sources of variation of resistance are not well established. Thus, baseline data are required in order to implement an efficient monitoring programme.

Documenting temporal trends in resistance patterns is an important objective of a monitoring programme. Nevertheless, effective prevention and intervention measures require more detailed knowledge of flock-level risk factors for the presence of resistance. The type of housing system is a known risk factor for infection of the flock with *Campylobacter*. The three predominant housing systems in Swiss poultry production are conventional indoor systems (floor keeping), indoor systems with access to an outdoor climate area (winter garden) and free range production with access to an outdoor area (pasture). Free range production has been associated with an increased risk of *Campylobacter* infection in the past (Choisat, 1997; Heuer et al., 2001). However, no information is currently available on whether there is an association between type of housing and the risk of antibiotic resistance.

The objectives of this research were to establish baseline data for the implementation of a monitoring programme for antibiotic resistance and to investigate the role of the housing system as a possible risk factor for antibiotic resistance. *Campylobacter* in poultry production was chosen as a model agent because it is the most frequent food-borne zoonotic infection in Switzerland.

MATERIALS AND METHODS

In the years 2002 and 2003, 100 poultry flocks per year were included in the study. Flocks were randomly selected from six producers who represent more than 95% of the Swiss poultry production. Samples were collected from March to May of each year because a medium prevalence of *Campylobacter* spp. was expected to be found during this time of the year. Five birds per flock were sampled at two sampling points before and after the slaughter process. Cloacal swabs were collected from live birds at delivery to the abattoir. Neck skin samples were taken at the end of the slaughter line. Samples were collected by meat inspectors and shipped to the laboratory overnight in a cooled container. In 2002, three flocks were lost to follow-up due to delayed shipping of the samples to the laboratory. In 2003, four flocks were missing from the analyses because one of the abattoirs failed to sample the selected flocks.

Samples were cultured for thermophilic *Campylobacter* spp. using standard bacteriological techniques. Each sample was inoculated into selective enrichment broth (*Brucella* broth [211088, Becton Dickinson, Sparks, USA] with *Campylobacter* growth and selective supplement [SR084E, SR069E, Oxoid, Basingstoke, Hampshire, UK]) and incubated at 42°C for 24h under microaerobic conditions provided by commercial gas packs (CampyGen from Oxoid). After enrichment, the samples were streaked on selective *Brucella* agar media (211086 Becton Dickinson) with 6% horse blood (SR0048C, Oxoid)) as well as Butzler *Campylobacter* selective supplement (SR085E, Oxoid) and incubated at 42°C for another 24h under microaerobic conditions. Translucent white, moist and glistening colonies were picked and taken for further identification. Identification of *Campylobacter* strains was performed using the standard bacteriological tests (gram-negative stain, characteristic motion, catalase and oxidase reactions, aerobic growth). For antibiotic resistance testing, the disc diffusion method recommended by National Committee for Clinical Laboratory Standards (NCCLS) was performed (National Committee for Clinical Laboratory Standards, 1997; 1998). The following antibiotic impregnated discs (bioMérieux SA, France) were used: erythromycin (15 μg), ciprofloxacin (5 μg), tetracycline (30 μg), gentamicin (10 μg), streptomycin (10 μg), ampicillin (10 μg), amoxycillin (25 μg) and sulphonamide (20 μg). Three to five isolated colonies of the same morphological type were selected from the agar plate culture and transferred into trypticase soy broth (211768, Becton Dickinson). After incubation at 42°C for 24h under microaerobic conditions, a sterile cotton swab was dipped into the suspension and streaked on the entire surface of a Mueller-Hinton agar (CM 337, Oxoid) with 5% sheep blood. Four antibiotic discs were placed on each plate and, after 48h of microaerobic incubation at 42°C, the diameter of the inhibition zone was measured with calipers. *E. coli* ATCC 25922 and *S. aureus* ATCC 25923 were used as reference strains. Zones of growth inhibition were evaluated according to the NCCLS standards. In addition, quantitative resistance data were obtained for ciprofloxacin, erythromycin and tetracycline using the E-test (AB Biodisk, Solna, Sweden). Inocula were prepared by incubating the strains for 24h at 42°C under microaerobic conditions in trypticase soy broth. After application of the E-test strips, plates were incubated in microaerobic conditions at 42°C for 48h. The minimal inhibition concentration (MIC) was read directly from the test strip at the point where the elliptical zone of inhibition intersected the MIC scale on the strip. The following NCCLS zone diameter (mm) and MIC breakpoints for resistance were applied: erythromycin $\leq$ 13 mm and MIC $\geq$ 8 mg/l, ciprofloxacin $\leq$ 15 mm and MIC $\geq$ 4 mg/l, tetracycline $\leq$ 14 mm and MIC $\geq$ 16 mg/l, streptomycin $\leq$ 11 mm, ampicillin $\leq$ 13 mm, gentamicin $\leq$ 12 mm, amoxycillin $\leq$ 13 mm and sulphonamide $\leq$ 12 mm.

Information on possible risk factors for resistance was collected by a questionnaire. Flock level risk factors taken into account were type of housing system, geographic location, flock size and breed. Possible confounding factors such as month of sampling, time of slaughter and production company were also recorded.

RESULTS

The prevalence of *Campylobacter* spp. differed among sampling points and between years. In the year 2002, 202 of 485 cloacal swabs (42%) and 106 of 485 neck skin samples (22%) were positive for *Campylobacter* spp.. In 2003, *Campylobacter* were isolated from 118 of 480 cloacal swab samples (25%) and from 65 of 480 neck skin samples (14%), respectively. In 76% of the positive samples, the isolated strains were identified as *Campylobacter jejuni*. *Campylobacter coli* was isolated from 23% of the samples while 1% of the samples contained both *C. jejuni* and *C. coli*.

Table 1. Prevalence of *Campylobacter* spp. in poultry samples from different origin.

	INDOOR PRODUCTION	WINTER GARDEN	FREE RANGE
Cloacal swabs 2002	43%	26%	54%
Neck skin 2002	25%	14%	27%
Cloacal swabs 2003	24%	18%	40%
Neck skin 2003	13%	10%	20%

In 2002, 20 of the 97 flocks included in the study originated from complete indoor housing (floor keeping), 34 from indoor housing with an outdoor climate area (winter garden) and 43 from free range production. Of 96 flocks sampled in 2003, 17 were produced in an indoor system, 51 were raised in a winter garden system and 26 were from free range production. For two flocks, the information on type of housing system was missing. The *Campylobacter* prevalence found in the different housing types in each year is presented in Table 1. The highest prevalence was found in free range systems, whilst indoor production with winter garden had the lowest prevalence. Production company and flock of origin were additional factors influencing the prevalence of *Campylobacter*. No effect of month of sampling was found for the three-month sampling period.

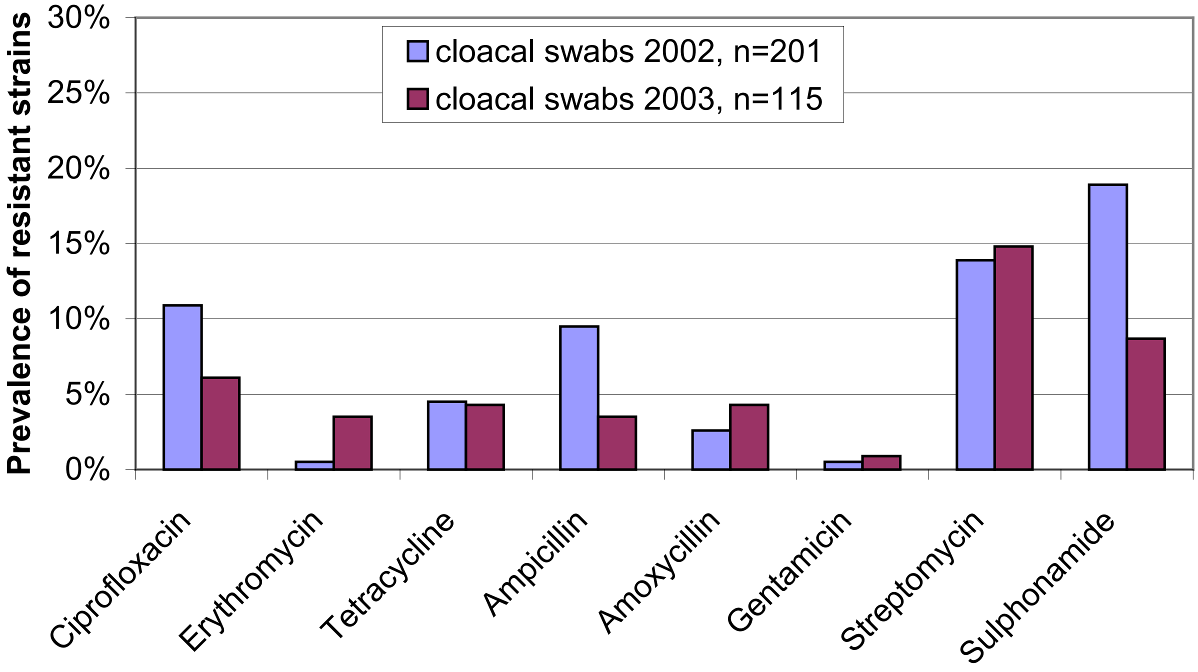


Fig. 1 Prevalence of resistance in *Campylobacter* isolates from cloacal swabs of live birds.

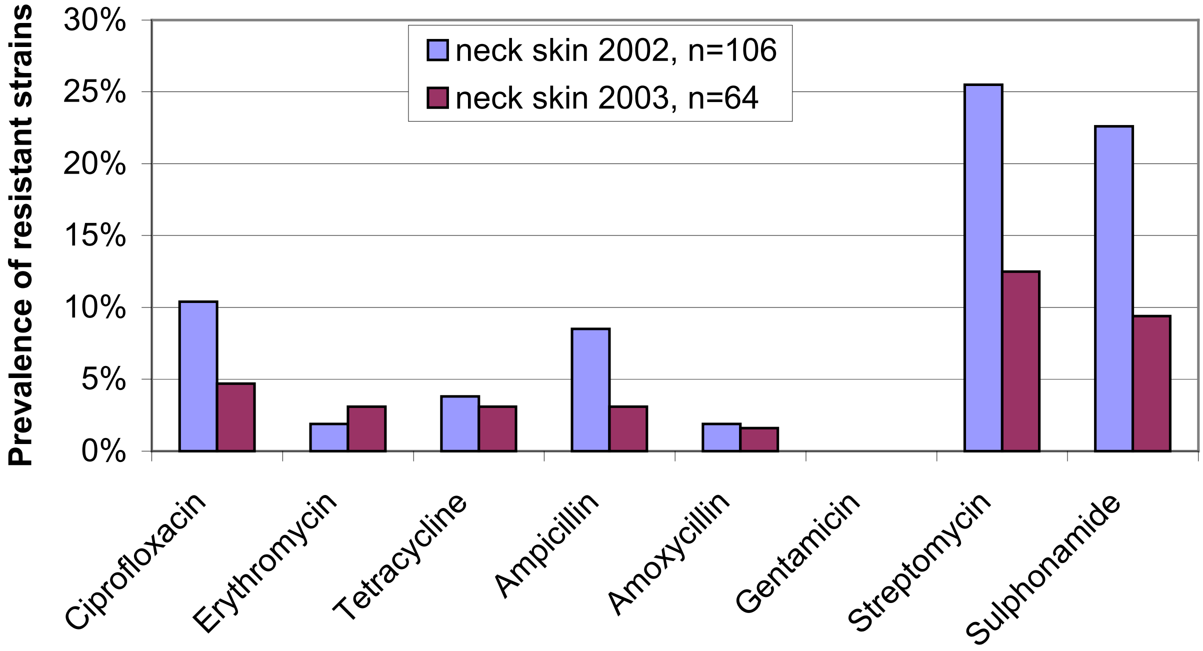


Fig. 2 Prevalence of resistance in *Campylobacter* isolates from neck skin samples.

Except for streptomycin resistance in 2002, there was no significant difference in the prevalence of resistance between *Campylobacter* isolates from cloacal swabs collected immediately after killing of the birds, and neck skin samples collected at the end of the slaughter line. Significant differences in the prevalence of resistance among study years were observed for ciprofloxacin, ampicillin and sulphonamides. Figures 1 and 2 show the prevalence of resistance against different antibiotics in *Campylobacter* isolates from cloacal swabs collected from live birds and from neck skin samples collected at the end of the slaughter line.

Resistance patterns differed among flocks raised in different housing systems. No tetracycline or erythromycin resistance was observed in *Campylobacter* strains from complete indoor production. *Campylobacter* isolates from production systems with winter garden had the highest prevalence of resistance against ciprofloxacin, erythromycin, tetracycline, streptomycin, and sulphonamides. In Fig. 3, the prevalence of resistance for the three most relevant antibiotics used for therapeutic treatment of human campylobacteriosis is shown for the different housing types. Significant differences between housing systems were found for ciprofloxacin and tetracycline resistance. *Campylobacter* strains isolated from winter garden systems were more likely to be resistant against more than one antibiotic than isolates from complete indoor farms and isolates from free range farms. The prevalence of multiple resistant strains in *Campylobacter* isolated from cloacal swabs was 9.5% for complete indoor farms, 30.7% for winter garden systems and 16% for free range systems, respectively. In addition to the effect of the housing system, there was a marked difference in resistance patterns among different production companies, as well as between individual flocks.

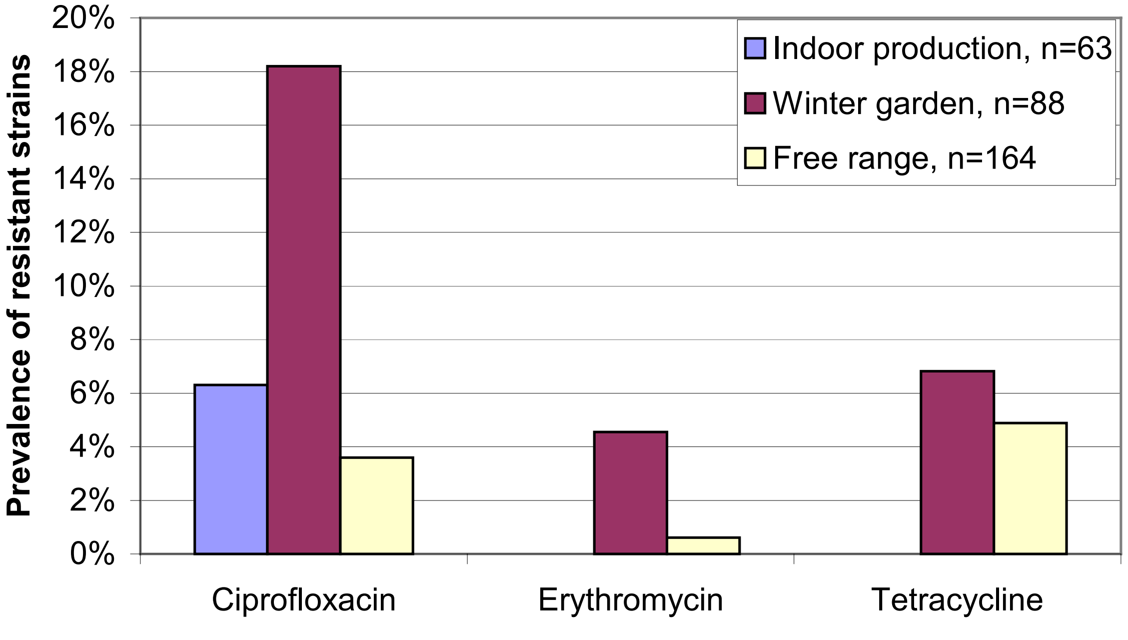


Fig. 3 Prevalence of resistance to ciprofloxacin, erythromycin and tetracycline in *Campylobacter* isolates from cloacal swabs from broilers produced in conventional indoor systems (floor keeping), in indoor production with an outdoor climate area (winter garden) and in free range outdoor production.

In 2003, *Campylobacter* resistant to more than two antibiotics were found in cloacal swabs from one flock only. From the five birds sampled from this flock, one *C. coli* isolate was resistant against four, and four *C. coli* isolates showed resistance against five antibiotics (ciprofloxacin, erythromycin, tetracycline, streptomycin and sulphonamide). Two *Campylobacter* strains with the same resistance pattern were isolated from the neck skin of birds from another flock slaughtered consecutively at the same abattoir. In 2002, six isolates from four flocks showed resistance to more than two antibiotics. One *C. coli* isolate was resistant against tetracycline, ampicillin, amoxycillin and sulphonamides. Two isolates each were resistant against the combination of tetracycline, ampicillin and amoxycillin and against the combination of tetracycline, streptomycin and sulphonamides. One isolate showed resistance against erythromycin, streptomycin and sulphonamides.

DISCUSSION

The prevalence of resistance observed in this study was relatively low. This is in agreement with another study which found a lower level of resistance to ciprofloxacin, tetracycline and ampicillin in poultry meat samples produced in Switzerland compared to imported meat sold in Switzerland (Ledergerber et al., 2003). Studies from Japan (Chuma et al., 2001), Spain (Saenz et al., 2000), and Austria (Anon., 2001) found considerably higher prevalences of antibiotic resistance than those observed in Swiss poultry. On the other hand, Denmark (Emborg et al., 2003) and Sweden (Bengtsson et al., 2003) reported lower levels of resistance against fluoroquinolones and tetracycline than those found in Switzerland.

In our study, a decreased prevalence of resistance against several antibiotics was observed in 2003 compared to 2002. The continuation of the monitoring programme in 2004 will show whether this decrease represents a temporal trend. According to veterinarians working in the Swiss poultry industry, 2003 has been a very good year in poultry production with few disease problems. However, it also cannot be excluded that the decrease in the prevalence of resistance was a bias introduced by the monitoring programme. All poultry production companies received their results compared to the overall results after the first study year. This may have prompted some companies to reinforce a more prudent use of antibiotics within their farms.

Resistance patterns of *Campylobacter* isolated from samples collected before and after the slaughter process were similar. Thus, for the continuation of the monitoring programme, it will be sufficient to concentrate sampling efforts to one of these two sampling points. Because the prevalence of *Campylobacter* was higher in cloacal swabs than in neck skin samples, this sampling point was more efficient for obtaining a precise estimate of the prevalence of resistance. An additional advantage of cloacal swab samples was that one major production company already routinely sampled flocks at this sampling point for a *Campylobacter* control programme. The prevalence of *Campylobacter* was lower in 2003 than in 2002. Most likely, this reflects the industry’s efforts to control *Campylobacter* infection. The lowest prevalence was observed in indoor systems with winter gardens. The *Campylobacter* control programme mentioned above focuses on this production type. In this programme, a premium price is paid for flocks that are found to be free of *Campylobacter* at slaughter. Other production companies have more informal programmes for the reduction of *Campylobacter*, which might have been less effective in reducing the *Campylobacter* load.

Although results have not yet been corrected for possible confounding factors such as month of sampling, time of slaughter and production company, preliminary analyses on risk factors for antibiotic resistance indicate a potential for improvement of the resistance situation by optimising the housing system. While housing systems with an outdoor climate area (winter garden) had the lowest prevalence of *Campylobacter*, the isolates from these systems showed the highest prevalence of antibiotic resistance. This may indicate that antibiotics were used in these systems in an effort to reduce the prevalence of *Campylobacter*. However, it is also possible that the relatively high prevalence of resistance is indicative of health problems associated with this type of production system. Data on the use of antibiotics in the study flocks were not available. Further research will be necessary to clarify these findings.

The information on prevalence of antibiotic resistance presented in this paper is being used as baseline data for the development of a Monte Carlo simulation model (Regula et al., 2003). This model will be utilised to optimise the cost and benefit of the monitoring programme for antibiotic resistance in Switzerland. Further development of the programme will include monitoring of other animal species, resistance testing of additional bacteria and production of national data on the usage of antibiotics.

ACKNOWLEDGEMENTS

This research has been funded by the Swiss Federal Veterinary Office and by the National Research Programme 49 ‘Antibiotic Resistance’ of the Swiss National Science Foundation.

REFERENCES

Anon. (1999). Resistance monitoring and surveillance. Report of a European Scientific Conference “The use of antibiotics in animals – Ensuring the protection of public health” organised by the World Animal Health Organisation (OIE) in Paris, March 24-26, 1999. www.antibiotics-conference.net

Anon. (2001). Resistance monitoring 2001. An area-wide study on resistance patterns of selected zoonotic pathogens, indicator bacteria, and bacteria causing mastitis in the Styrian food animal population. (in German) REMOST 2001, Fachabteilung 8C-Veterinärwesen, Amt der Steiermärkischen Landesregierung, Graz, Austria.

Bengtsson, B., Greko, C. and Wallen, C. (2003). SVARM 2002. Swedish veterinary antimicrobial resistance monitoring. Department of Antibiotics, National Veterinary Institute, Uppsala, Sweden.

Caprioli, A., Butani, L., Martel, J.L. and Helmuth, R. (2000). Monitoring of antibiotic resistance in bacteria of animal origin: epidemiological and microbiological methodologies. Intern. J. Antimicrob. Agents 14, 295-301

Choisat, B. (1997). The epidemiology of *Campylobacter jejuni* in poultry farms in Switzerland. (in French) Doctoral thesis, University of Berne, Faculty of Veterinary Medicine, Bern, Switzerland.

Chuma, T., Ikeda, T., Maeda, T., Niwa, H. and Okamoto, K. (2001). Antimicrobial susceptibilities of *Campylobacter* strains isolated from broilers in the southern part of Japan from 1995 to 1999. J. Vet. Med. Sci. 63, 1027-1029

Dunlop, R.H., McEwen, S.A., Meek, A.H., Friendship, R.M., Black, W.D. and Clarke, R.C. (1999). Sampling considerations for herd-level measurement of faecal *Escherichia coli* antimicrobial resistance in finisher pigs. Epidemiol. Inf. 122, 485-496

Emborg, H.D. and Heuer, O.E. (2003). DANMAP 2002 – Use of antimicrobial agents and occurrence of antimicrobial resistance in bacteria from food animals, foods and humans in Denmark. Danish Veterinary Institute, Copenhagen, Denmark.

Heuer, O.E., Pedersen, K., Andersen, J.S. and Madsen, M. (2001). Prevalence and antimicrobial susceptibility of thermophilic *Campylobacter* in organic and conventional broiler flocks. Lett. Appl. Microbiol. 33, 269-274

Ledergerber, U., Regula, G., Stephan, R., Danuser, J., Bissig-Choisat, B. and Stärk, K.D.C. (2003). Risk factors for antibiotic resistance in *Campylobacter* spp. isolated from raw poultry meat in Switzerland. BMC Public Health 3, 39

National Committee for Clinical Laboratory Standards (1997). Performance Standards for Antimicrobial Disc Susceptibility Tests. 6th edn. Approved standards M2-A6. Wayne, National Committee for Clinical Laboratory Standards, 1-17.

National Committee for Clinical Laboratory Standards (1998). Performance Standards for Antimicrobial Susceptibility Testing. 8th informational supplement M100-S8. Wayne, National Committee for Clinical Laboratory Standards, 14-15.

Regula, G., Lo Fo Wong, D., Stephan, R., Bissig-Choisat, B., Danuser, J., Ledergerber, U. and Stärk, K.D.C. (2003). Optimization of an antimicrobial resistance monitoring program, using *Campylobacter* spp. in poultry production as an example. Proceedings 10th International Symposium for Veterinary Epidemiology and Economics, November 17-21, 2003, Vina del Mar, Chile

Saenz, Y., Zarazaga, M., Lantero, M., Gastanares, M.J., Baquero, F. and Torres, C. (2000). Antibiotic Resistance in *Campylobacter* strains isolated from animals, food, and humans in Spain in 1997-1998. Antimicrob. Agents Chemother. 44, 267-271

Wray, C. and Gnanou, J.C. (2000). Antibiotic resistance monitoring in bacteria of animal origin: analysis of national monitoring programmes. Intern. J. Antimicrob. Agents 14, 291-294

ANIMAL HEALTH ECONOMICS

THE STATE OF ANIMAL HEALTH ECONOMICS: A REVIEW

K.S. HOWE* AND K.H. CHRISTIANSEN

SUMMARY

The importance of investigating the economic impacts of health problems in animal populations is widely acknowledged. However, a literature review leaves mixed impressions of the nature and scope of research in animal health economics. There is evidence of significant progress, but there has also been a tendency to overlook the need to correct the definition or interpretation of certain economic concepts and relationships used. Moreover, there are no logical grounds for regarding animal health economics as a discipline. Neither is it a collection of techniques. Viewed as such, it will fail to develop an analytical framework to guide decisions for a modern problem agenda in which economy-wide disease impacts, including food safety issues, are of pre-eminent concern.

INTRODUCTION

Since the 1970s, analysing the economic implications of problems in animal health has become a prominent feature of policy-oriented research. Dedicated post-experience courses are now delivered worldwide, and Masters degrees in veterinary epidemiology invariably include an economics component. The authors both teach and research in the field of animal health economics, and this paper is partly the outcome of their reflections on concerns arising from that experience. It is also a response to calls from colleagues in the Society for Veterinary Epidemiology and Preventive Medicine to give more prominence in the Society's Proceedings to economic aspects of problems in animal health. That the subject is deemed important enough to justify more discussion is sufficient reason to ask some fundamental questions about it.

OUR APPROACH

The majority of papers delivered at the Society's conferences and published in its Proceedings report the outcome of specific items of empirical research. This paper is different. It has the express purpose of provoking reflection and reactions across a wide range of methodological issues. Therefore, the broad perspective on animal health economics taken here is deliberate. An aim is to contribute to the further development of animal health economics as a framework for guiding resource use decisions in relation to animal health. Also, as becomes increasingly important, it is necessary to have regard for the relationships between problems in animal and human health.

---

* Keith Howe, Centre for Rural Research, University of Exeter, St German's Road, Exeter, Devon, EX4 6TL, UK.   E-mail: K.S.Howe@exeter.ac.uk

As we argue below, there is ample reason for concern that currently all is not as it should be. The discussion is informed by a literature review. In no sense, though, was this comprehensive. The objective was to pinpoint contributions to the literature which illustrate either the main directions for research in animal health economics or innovative approaches. Moreover, the majority of publications reviewed are from journals. Thus economic studies of animal health issues typically published by the commissioning organisations, for example government departments, are mostly excluded from consideration here. Certainly a more extensive review is merited.

If any criticism levelled below is refuted by a publication or publications inadvertently omitted, so much to the good. Our objective is only to investigate whether animal health economics is broadly found to be on a sound scientific footing. In the course of their teaching and research, the authors have noted contributions to the animal health economics literature thought worthy of note and quotation. Many are examples of 'good practice', but often more interesting are those which are not, or at least not categorically so. Patterns can be discerned which are reflected in the structure of the following discussion. Remarkably, it is necessary to begin with an issue which ought to have been laid to rest long ago.

ANIMAL HEALTH ECONOMICS IS NOT A DISCIPLINE

Curiously, even more recent literature claims that animal health economics is a relatively new discipline. For example, Dijkhuizen, Huirne and Jalvingh (1995, page 135) state that "Animal Health Economics is a relatively new discipline, which is progressively developing a solid framework of concepts, procedures, and data to support the decision-making process in optimizing animal health management." Perry and Randolph (1999, page 165) similarly conclude that " a new discipline of animal health economics has emerged". Yet there is no convincing basis for the assertion. For example, in his exploration of 'forms of knowledge' Hirst (1963) defines a discipline according to four criteria. The first of these is that "They each involve certain central concepts that are peculiar in character to the form. For example, those of gravity, acceleration, hydrogen, and photosynthesis characteristic of the sciences; number, integral and matrix in mathematics; God, sin and predestination in religion; good and wrong in moral knowledge."

On this criterion alone, animal health economics fails as a discipline. It is subservient to economics which does indeed have its own central concepts such as value, the firm, markets, and social welfare. All of these concepts are mobilised in relation to analysing decisions for achieving resource use efficiency in animal health. Animal health economics more closely approximates to what Hirst defines as a "field", i.e., " those organisations [of knowledge] which are not themselves disciplines or subdivisions of any discipline. They are formed by building together round specific objects, or phenomena, or practical pursuits, knowledge that is characteristically rooted elsewhere in more than one discipline."

There are significant further implications of Hirst's definitions. Arguably, veterinary epidemiology qualifies as a discipline. It draws on various branches of knowledge, notably the biological sciences, mathematics and statistics, to address problems of disease in animal populations. But the outcome is an analytical framework which is more than the sum of its individual parts. Concepts such as prevalence, incidence, populations at risk, endemic and epidemic, point to its own distinctive features, and thus disciplinary credentials.

Part of economics makes its contribution to epidemiology, too, since disease in animal populations creates value losses to human society because of reduced output and the diversion of resources with positive opportunity costs to disease control. Thus the conclusion reached by Howe (1989) that making an explicit distinction between economics and veterinary epidemiology (for example, as in the title International Symposium for Veterinary Epidemiology and Economics) is illogical is reaffirmed from another perspective.

ECONOMICS AND BIOLOGICAL PROCESSES

The strictly economic dimensions to disease in animal populations are all handled perfectly well by that discipline's own standard apparatus of concepts and theory. It does not make sense to claim that 'animal health economics' is somehow different from conventional economics when applied to animal disease problems, and that it offers the prospect of novel insights. Precisely the same arguments apply to agricultural economics which, as some explicitly recognise (e.g., Perry & Randolph, 1999), has close affinities with animal health economics. The common ground is that both focus on aspects of the efficiency of resource allocation and transformation for human well-being in a context dominated by biological relationships.

From a technical point of view, analysis of biological systems can get very complicated. This applies, for example, to the dynamics of changes in animal populations, in which a production process takes place over multiple time periods, and involves complex relationships between investment and consumption. Yet the roots of such particular complexity are in the technical relationships, not the economics. In no sense do the fundamental concerns of economics become transformed by the peculiarities of the physical world. As always, the technical relationships set the constraints within which economic decisions have to be made.

THE ECONOMICS OF DISEASE, NOT HEALTH

Conceptual distinctions which are not absolutely crucial should not have their importance overemphasised. Arguably, the designation 'animal health economics' as opposed to 'economics of animal disease' falls into that category. However, in anticipating the discussion of more specific considerations which follow, it is useful to pause and reflect on the wider ramifications of the choice of designation which has become the universal convention.

Health is not a problem; disease is. Similarly, a world in which a multitude of producers has perfect knowledge of the most up-to-date technology, unlimited access to all the resources they require for production at the lowest possible cost, complete knowledge of everything their many competitors are doing, and all consumers know where to get what they want, when they want it, at minimum price, is not a problem. In fact, the economic world described - recognisable to any economics student as a crude description of the market structure known as 'perfect competition' - is an ideal. It corresponds to a situation in which people's overall economic welfare is maximised. And, of course, as is true by definition for any ideal, it is a situation that does not exist in reality.

Nevertheless, the ideal provides us with a benchmark against which to assess the consequences of any change in the economy; quite simply, does any change move people's well-being closer to achieving the ideal outcome, or away from it? The fact that perfection is unattainable is beside the point. What matters is getting closer to it. So it is with animal disease. We cannot realistically envisage a world with zero disease, but we can, and do, take measures to mitigate its adverse effects, i.e. to change an existing situation (disease in animal populations) into something preferred (less disease in animal populations).

It follows that our focus as economists in animal populations is strictly disease, not health. From an analytical point of view, the perspective is important. For presentational purposes 'animal health economics' will suffice unless it leads to substantive misunderstanding of what the field is all about. Sometimes it does. Moreover, lack of precision may contribute to other, more specific, 'rough edges' to which discussion now turns.

PRINCIPLES BEFORE TECHNIQUES

That much progress has been made in the field of animal health economics is beyond dispute. Unfortunately, some bad habits have been acquired along the way. For example, Morris (1999 page 305) begins, incorrectly, by describing economic analysis as "a valuable technique". It is not a technique. Economic analysis rests on a formidable array of concepts and theory which, flexible in their application, help us to disentangle the most complex empirical relationships impinging on people's (mainly) material well-being. It does so by the application of a rigorous and logical framework, honed and refined over some two centuries, to a point where 'powerful simplicity' is a justifiable summary of economics' contribution as a social science.

Yet animal health economics undoubtedly retains a focus on the application of techniques or, to be precise, a relatively narrow range of techniques. Much empirical work in the field begins with some broad observations about the nature of the economic context, and then leaps into consideration of techniques. The review article by Dijkhuizen et al. (1995) is a case in point. So, most notably of all, is Dijkhuizen and Morris (1997) which, although it purports to be a text about animal health economics, is hardly about economics at all.

Morris (1999) asserts that five common methods of economic analysis are applied in the evaluation of animal health issues. These are a) partial budgeting, b) enterprise analysis, c) cost-benefit analysis, d) decision analysis, and e) system modelling. Morris points out that these can be used in combination as the particular problem context demands. His classification differs from those of Dijkhuizen et al., in turn quoted by Disney et al. (2001), only by the inclusion of enterprise analysis. In fact, these are an 'ad hoc' mixture of techniques and structured approaches to problem analysis. The authors are actually in search of a set of recipes, a kind of 'cook book' of procedures, to facilitate particular aspects of economic analysis for everyday purposes. That has its role, but it is achieved at the considerable risk that any student who comes to acquire some knowledge of economics from study of those procedures is likely to grab the wrong end of a very long stick.

HEADING DOWN A 'CUL DE SAC'

Let us be precise about the target of our criticism. Obviously, empirical analysis always requires the use of technique(s); the one goes inextricably with the other. The danger lies in being seduced into adopting an analytical approach which starts off by looking for ways to package any given economic problem into something that fits a particular technique or techniques (or 'method'). Even on the principle of 'safety first', the logical approach is to begin by thinking around an economic problem in an unconstrained way, and only then deciding on how best to approach its empirical analysis. Otherwise, economic analysis comes to be regarded as a choice between algorithms. More often than not, economic problems cannot be treated like that.

For sure, there will be instances of classes of economic problems with common structural characteristics. Therefore, they are amenable to application of the same techniques. In a particular sense, therefore, they cease to be especially interesting as exercises in economic analysis because they are simply routine. The interest is entirely, and very properly, in the results. But that is beside the crucial point which bears re-emphasis. The point at issue concerns maintaining a proper perspective on the methodology of economic analysis. Without it, there is a real danger that the ongoing development of animal health economics will end in an intellectual 'cul de sac'. In other words, packaging animal health problems in such a way that they come to be seen as classifiable into just four or five types for routine economic analysis denies the richness of economics as an adaptable framework for illuminating a much wider range of important issues.

Thus Ramsay et al. (1999, page 343) are correct to say that "economic analysis in animal health has generally been undertaken within an animal health framework that has limited the application of the wider methodologies provided by economists." Similarly, when Morris (page 306) states that "an extensive body of published examples of analyses in animal health economics is now available; these can act a models, covering almost all of the types of evaluation issues likely to be faced by analysts", great care is appropriate. Analysis informed by case studies puts analysts at risk of overlooking any unique features of the current problem context which may have a crucial bearing on research outcomes.

COST-BENEFIT ANALYSIS

It is impossible to be in the company of a veterinary scientist who has even the most cursory acquaintance with economics without the words 'cost-benefit analysis' soon being mentioned. Indeed, at one time cost-benefit analysis was virtually synonymous with animal health economics. There are numerous examples of such studies, including James (1987), Ansell (1988), Berentsen et al. (1992a, 1992b), Anaman et al. (1994), Aubert (1999), Perry and Randolph (1999), Tambi et al. (1999), Valle et al. (2000), Disney et al. (2001) and James & Rushton (2002),

In one sense, much of economics is about cost-benefit analysis. The allocation of scarce resources always involves making decisions about what to use them for, and in what quantities. Benefits are created, but also costs because output(s) which might have been obtained from using the resources differently are consequently foregone, i.e. there are opportunity costs. However, this is not what is meant by cost-benefit analysis in its more specific definition.

Strictly defined, *social* cost-benefit analysis concerns the appraisal of projects which have social impacts, irrespective of whether they are undertaken for private or public reasons (Campbell & Brown, 2003). Sugden and Williams (1978) distinguish clearly between financial appraisal and social cost-benefit analysis, while Mishan (1988) provides a detailed exploration of the approach's theoretical foundations and breadth of application.

Ramsay et al. (1999) are unusual in the animal health economics literature for explicitly drawing attention to the important distinction between cost-benefit and discounted cash flow analysis, which focuses on financial flows over time. There is a tendency in the literature for the two to be treated as synonymous (e.g., Rushton et al. 1999;Tambi et al., 1999). Formally, discounted cash flow analysis constitutes part of cost-benefit analysis, but of itself does not account for the adjustments to prices, valuation of externality effects, or corrections for price distortions resulting from trade barriers, which are important matters in social cost-benefit analysis properly defined.

In short, any analysis which involves a comparison over time of benefits and costs, whether strictly financial or otherwise, and therefore requires discounting is commonly, if erroneously, deemed cost-benefit analysis. In practice, such conceptual confusion may not be critical in empirical work. All the same, it is unnerving to be confronted with evidence that researchers sometimes build their analysis on a less secure foundation of understanding than it is reasonable to expect.

It is also important to reflect on the strengths and limitations of cost-benefit analysis as a source of information for decisions. For example, obviously it makes sense 'ex ante' to appraise whether a disease control programme under consideration is likely to produce positive net social benefits, i.e., a benefit-cost ratio of at least unity. On the other hand, 'ex post' project appraisal can only show whether or not a mistake was made. If, say, it is found that the expected benefit-cost ratio from a disease control programme already being implemented is less than unity the options are a) stop the programme immediately to avoid 'wasting' still more resources, b) find ways to modify the existing programme in ways which then bring a positive expected net benefit flow over time, or c) take the adverse result into account when considering implementation of similar disease control programmes in other locations.

Furthermore, cost minimisation is a key criterion for economic efficiency. As McInerney (1991) shows, a high benefit-cost ratio typically will not identify the control strategy which minimises the total costs of disease. Thus cost-benefit analysis is likely to give misleading results if comparison of the magnitude of benefit-cost ratios for alternative disease control programmes is used as the sole criterion for choice of programme.

ELEMENTARY ERRORS

The above discussion points to some of the hazards of applying the tools of economic analysis when their interpretation is imperfectly understood. Morris (1999) states that "Those new to the techniques often fear making errors in analyses (which will be detected by experts with differing backgrounds from their own), and also fear using the terminology of the field incorrectly." To which the only response is, and so they should! Presumably the unspecified 'experts' are economists who validly expect research results to be based on application of their discipline with the same intellectual rigour as would any other scientist of their own.

However, other much more basic errors occur in the literature: First, staying with Morris (1999, page 312), it is simply not true that "economics is built around the concept of the production function." Important as the concept is, it relates to technical relationships on the supply side, mainly in microeconomics. It has nothing to do with demand or most of macroeconomics, complete sub-fields of economics in themselves.

Second, the term 'macroeconomic' is itself invariably defined unconventionally in animal health economics, where it is taken to be synonymous with sector level issues in microeconomics (see, for example, Dijkhuizen et al., 1995, page 145). In its conventional usage, to be discovered from any basic economics textbook (e.g., Begg et al., 1997), its focus is mainly inflation, unemployment, and economic growth, by definition in the context of the whole economy.

Third, even formal economic models presented in diagram form can be subject to cavalier treatment. For example, in Dijkhuizen et al. (also page 145) and, with a slightly different explanation, Berentsen et al. (1992a), attention is validly directed to the importance of the market interaction of supply and demand. But then a figure is presented which shows the situation for a country exporting a product. The amount exported at a 'basic price level' (whatever that means) is shown, and the consequences of an export ban interpreted in terms of "a new equilibrium at a lower price level." The definition of market equilibrium is precise and unambiguous in economics. It refers to the particular combination of a unique price and a corresponding quantity of product at which there is neither excess demand nor excess supply. In other words, in a particular sense, consumers and producers simultaneously are content; consumers are not left wanting more at the going price, and producers sell everything they produce.

So, if a country is an exporter at a given price, its own domestic market is not in equilibrium and, by definition, the aforesaid 'basic price level' is not a (domestic) equilibrium price. It may indeed be the equilibrium price for that product on the world market, which is altogether a different thing. That is determined by the simultaneous interaction of all trading countries' supply and demand schedules. Consequently, an export ban in one country may, or may not, cause the world equilibrium price to fall. It all depends on the share of that country in world trade. If, as the USA for example, it is a major player, whatever the country does in its domestic policy will have economic effects (price and quantity) transmitted world-wide. If it is a small country, the effects of its own actions may be imperceptible.

What matters here is not so much the particular examples quoted, but the general principle being illustrated. If animal health economics is to progress as an aid to decision-making for animal disease control, it needs much greater input from mainstream economics and economists. This would help to eliminate the kind of inaccuracies highlighted. True, sometimes they are unimportant in relation to analytical outcomes. After all, it is not critical to know that economics is not built around the concept of a production function as long as the concept is used appropriately. On the other hand, sometimes precision does matter, as in the interpretation of international market effects.

To a large extent, animal health economics to date has been dominated by the contributions of veterinary epidemiologists who have embraced economics, and farm economists. Without their indispensable contribution, the economic dimensions of problems in animal disease probably would not have been recognised. 'Mainstream' economists, by and large, have shown remarkably little interest in applying their tools to problems in animal disease. But by ceding a virtual monopoly of the field to 'traditional' animal health economists, other economists have abandoned ground which more properly belongs to them, and with detrimental effects. Both of these groups, whose work is in fact complementary, therefore bear a share of the responsibility for shortcomings currently discernible in animal health economics.

EVIDENCE OF PROGRESS

Despite the reservations expressed above, there is also evidence of much progress having been made in problem definition and analysis in animal health economics.

Welfare economics

One particularly noteworthy change has been increasing recognition that animal disease and its control has variable impacts on different groups in human society, and on society as a whole. The economic policy analyst's standard framework of welfare economics, commonly referred to in the animal health economics literature as the 'economic surplus model', has become a familiar tool. Whereas Howe (1992) could cite only Amosson et al. (1981), Jetter and Klein (1984), and Power and Warwick (1984), examples now to be added include Ebel et al. (1992), Anaman et al. (1994), Ott et al.(1995), Kristjanson et al. (1999), and Mangen and Burrell (2003).

Of the later papers, Ebel et al. (1992) allow for the effects of changes over time in supply elasticity on producer surplus in the context of a national pseudorabies eradication program. Anaman et al. build on that approach, integrating welfare and social cost-benefit analysis, all based on a technically very detailed dynamic spatial economic model. On a different level of complexity, Ott et al.'s assessment of the national benefits from reducing livestock mortality is a good starting point for getting acquainted with the essential basics of empirical economic welfare analysis.

Kristjanson et al. (1999) is another extremely thorough study. Measuring the economic consequences of African animal trypanosomosis links geographical information systems to a biophysical herd simulation model and an economic surplus model. The adoption rate, and therefore diffusion curves, for implementing new control procedures is explicitly taken into account, and benefit/cost ratios computed for returns to vaccine research. The similarly thorough Mangen and Burrell (2003) study combines a sector-level partial equilibrium simulation model with a stochastic epidemiological model to investigate the welfare effects of classical swine fever epidemics in the Netherlands during 1997-98.

Characteristic of all these studies is the way in which detailed technical relationships are combined with the standard apparatus of economics to generate empirical results. Also, it is worth noting that three of the five studies mentioned particularly appear in other than veterinary journals. Clearly, for reasons of both sample size and selection bias, care must be taken not to make too much of this. However, it may be of some significance that issues in animal health are now being explored in more mainstream, especially economics, literature.

System effects

Another, more novel, aspect of certain empirical studies is the use made of Leontief input-output models. For example, Garner and Lack (1995) evaluated alternative control strategies for foot-and-mouth disease (FMD) in Australia. Their detailed technical analysis and stochastic disease simulation model is supplemented by regional input-output analysis to estimate the income and employment multiplier effects of FMD outbreaks. Also in relation to FMD, Mahul and Durand (2000) investigate the impact of international trade restrictions both on the export sectors directly affected and on the French economy as a whole. They also speculate on the desirability of using a computable general equilibrium model as an alternative approach. That would enable the price and quantity adjustments which ensure equilibrium in all markets to be estimated, which the input-output approach cannot do.

Such examples as these emphasise the way in which problems in animal health are recognised as being embedded in the wider economy, and therefore have knock-on effects far beyond the animal production sectors. This recognition, above all, places animal health economics in its proper place as a sub-field of general applied economics. Certainly those epidemiologists and animal health economists who work in developing countries, a number of whom have been quoted here, will be fully aware that their work is of national economic importance. By definition, developing country economies are highly dependent on agriculture and other primary commodity production and trade. Thus the productivity of livestock sectors in such countries is a significant consideration in people's economic well-being.

Modernising the problem agenda

The paper by Majorowski et al. (2001) on echinococcosis in Tunisia is a particular illustration of the widespread economic impacts of a preventable zoonosis in a developing country. But, as everyone knows, the effects of non-human animal disease on the human population, whether or not directly as a zoonosis, is now a major issue even in high income developed countries. Evidence for this is, for example, Cohen et al. (1999) who investigate the cost-effectiveness of bovine spongiform encephalopathy (BSE) screening. In the context of traceability as part of disease control, Disney et al. (2001) make an economic appraisal of animal identification systems. Lloyd et al. (2001) analyse the impact of food scares on price adjustment in the UK beef market, and Jin and Koo (2003) take a revealed preference approach to analysing structural change in consumption patterns resulting from BSE in Japan. Also concerned with public responses to BSE, Sanjuán and Dawson (2003) apply cointegration techniques to the study of producer and retail price transmission for beef, lamb and pork in the UK.

In all such instances, the motivation for identifying and measuring the economic impacts of animal disease are at least one step removed from animal production itself. Concerns range from the implications of changes in consumer preferences for the structure of animal production to, perhaps most important of all, how people perceive the effects of animal disease on their own health and well-being, and what they do about it. The entire area of food safety merits inclusion in the problem agenda for animal health economists, thus taking them beyond their traditional preoccupations. Moreover, as the public response to the UK foot-and-mouth crisis in 2001 showed, the indirect effects of disease outbreaks can have significant adverse effects on both urban and rural local economies, especially those dependent on tourism. A country's foreign exchange earnings, and thus balance of payments, are also affected. Indeed, the indirect economic effects may well outweigh the direct effects of animal disease by a significant margin. The economic analysis of animal disease has always been driven by concern to find efficient solutions to actual problems. Now that the problem agenda is changing, so must economists' focus in response.

CONCLUSIONS

This review of the state of animal health economics, admittedly limited, nevertheless leads to clear conclusions.

First, there is ample evidence of some ingrained bad habits in the way animal health economics is understood. It is not a discipline, and expectations that the field will evolve its own, unique, " solid framework of concepts, procedures, and data" (see above) always will be frustrated. There is only the complexity of biological systems in animal populations, the unpredictability of things that go wrong with them (disease), and the various tools and methods of human interventions to make corrections when they do. Mathematical modelling and statistical analysis, traditionally core instruments of epidemiology, are indispensable for discerning the systematic relationships which underlie the behaviour of such systems, disguised as they often are by other variables which appear as random effects.

In turn, economics is about finding ways to maximise people's well-being from using scarce resources as efficiently as possible. Animal disease causes lost output and diversion of resources which might have been used for beneficial purposes other than disease control. Both lost output and diverted resources therefore diminish people's well-being. The biological/technical relationships in disease and its control define the constraints within which economic decisions are made, and economics provides the rigorously defined efficiency criteria to guide such decisions. In short, everything necessary for economic analysis in relation to animal disease is already in place. On that score, there is nothing more to say. What matters is that the standard apparatus is understood and applied correctly.

Second, the importance of veterinary epidemiologists turning their attention to analysing the economic consequences of animal disease cannot be overestimated. Notwithstanding any criticisms levelled above, a relatively small number of authors, Morris and Dijkhuizen most prominent amongst them, unarguably have made a major and indispensable contribution. By contrast, the lack of interest of economists as such is conspicuous by the relative absence of their contributions. Unfortunately, that lack of interest has also tended to restrict problem definition and the choice of analytical approaches. That situation shows signs of changing, driven as much as anything by new concerns for food safety, often associated with the perceived consequences of intensive agriculture, and international trade liberalisation.

Third, and finally, the literature reviewed as the basis for this paper shows evidence of a very mixed appreciation of economic analysis. Animal health economics is still dominated by a tendency to see everything as a question of choice of technique(s). No doubt this is because of its role under the umbrella of veterinary epidemiology which, necessarily, has a focus on techniques. Yet this is to misunderstand the purpose, nature and scope of economic analysis. There is still need for a textbook of animal health economics which addresses these themes, and emphasises the concepts and relationships of economic theory rather than techniques.

REFERENCES

Amosson, S.H., Dietrich, R.A., Talpaz, H. and Hopkin, J.A. (1981). Economic and epidemiologic policy implications of alternative bovine brucellosis programs. West. J. Agric. Econ. 6, 43-56

Anaman, K.A., Atzeni, M.G., Mayer, D.G. and Stuart, M.A. (1994). Benefit-cost analysis of the use of sterile insect technique to eradicate screw worm fly in the event of an invasion of Australia. Prev. Vet. Med. 20, 79-98

Ansell, D.J. (1988). Research into bovine parasitic bronchitis: an economic appraisal. In Ansell, D.J., Done, J.T. Veterinary research and development: Cost-benefit studies on products for the control of animal diseases. Centre for Agricultural Strategy Joint Publication No 3, University of Reading, 27-46.

Aubert, M.F.A. (1999). Costs and benefits of rabies control in wildlife in France. Rev. Sci. Tech. Off. Int. Epiz. 18, 533-543

Begg, D., Fisher, S. and Dornbusch, R. (1997). Economics. 5th edition. McGraw-Hill, Part 4

Berentsen, P.B.M., Dijkhuizen A.A. and Oskam, A.J. (1992a). A critique of published cost-benefit analyses of foot-and-mouth disease. Prev. Vet. Med. 12, 217-227

Berentsen, P.B.M., Dijkhuizen, A.A. and Oskam, A.J. (1992b). A dynamic model for cost-benefit analyses of foot-and-mouth disease. Prev. Vet. Med. 12, 229-243

Campbell, H. and Brown, R. (2003). Benefit-Cost Analysis: Financial and Economic Appraisal Using Spreadsheets. Cambridge University Press. 343pp

Cohen, C.H., Cesbron, J-Y. and Valleron, A-J. (1999). Cost-effectiveness of bovine spongiform encephalopathy screening. Vet. Rec. 44, 703-706

Dijkhuizen, A.A., Huirne, R.B.M. and Jalvingh, A.W. (1995). Economic analysis of animal diseases and their control. Prev. Vet. Med. 25, 135-149

Dijkhuizen, A.A. and Morris, R.S. (1997). Animal Health Economics: Principles and Applications. University of Sydney. 306pp

Disney, W.T., Green, J.W., Forsythe, K.W., Wiemers, J.F. and Weber, S. (2001). Benefit-cost analysis of animal identification for disease prevention and control. Rev. Sci. Tech. Off. Int. Epiz. 20, 385-405

Ebel, E.D., Hornbaker, R.H. and Nelson, C.H. (1992). Welfare effects of the national pseudorabies eradication program. Am. J. Agric. Econ. 74, 638-645

Garner, M.G. and Lack, M.B. (1995) An evaluation of alternate control strategies for foot-and-mouth disease in Australia: a regional approach. Prev. Vet. Med. 23, 9-32

Hirst, P.H. (1963). Liberal education and the nature of knowledge. In Archambault, R.P. (ed.) Philosophical Analysis and Education. Routledge and Kegan Paul. London; reprinted in Brown, S., Fauvel, J., and Finnegan, R. (1981) Conceptions of Inquiry. Methuen with the Open University Press, 230-233

Howe, K.S. (1989). Conceptual affinities in veterinary epidemiology and economics. Acta Vet. Scand., Supplementum 84, 347-349

Howe, K.S. (1992). Epidemiologists' views of economics - an economist's reply. Proceedings of the Society for Veterinary Epidemiology and Preventive Medicine, Edinburgh, 157-167.

James, A.D. (1987). Principles and problems of benefit-cost analysis for disease control schemes. In Howe, K.S., McInerney, J.P.M. (eds.) Disease in Farm Livestock: Economics and Policy, Commission of the European Communities, 69-78

James, A.D. and Rushton. J. (2002). The economics of foot and mouth disease. Rev. Sci. Tech. Off. Int. Epiz. 21, 637-644

Jetter, F.P. and Klein, K.K. (1984). Economic benefits from the Alberta Warble Control Program. Paper to the annual meeting of the Canadian Agricultural Economics and Farm Management Society, University of Lethbridge, Alberta, 20pp

Jin, H.J. and Koo, W.W. (2003). The effect of the BSE outbreak in Japan on consumers' preferences. Eur. Rev. Agric. Econ. 30, 173-192

Kristjanson, P.M., Swallow, B.M., Rowlands, G.J., Kruska, R.L. and de Leeuw, P.N. (1999). Measuring the costs of African animal trypanosomosis, the potential benefits of control and returns to research. Agric. Sys. 59, 79-98

Lloyd, T., McCorriston, S., Morgan, C.W. and Rayner, A.J. (2001). The impact of food scares on price adjustment in the UK beef market. Agric. Econ. 25, 347-357

Mahul, O. and Durand, B. (2000). Simulated economic consequences of foot-and-mouth disease epidemics and their public control in France. Prev. Vet. Med. 47, 23-38

Majorowski, M.M., Carabin, H., Kilani, M. and Bensalah, A. (2001). Echinococcosis in Tunisia: an economic analysis. Proceedings of the Society for Veterinary Epidemiology and Medicine, Noordwijkerhout, The Netherlands, 187-201.

Mangen, M-J.J. and Burrell, A.M. (2003). Who gains, who loses? Welfare effects of classical swine fever epidemics in the Netherlands. Eur. Rev. Agric. Econ. 30, 125-154

McInerney, J.P. (1991). Cost-benefit analysis of livestock disease: a simplified look at its economic foundations. Proceedings of the 6th International Symposium on Veterinary Epidemiology and Economics, 149-153.

Mishan, E.J. (1988). Cost-Benefit Analysis. Unwin Hyman. London. Fourth edition. 461pp

Morris, R.S. (1999). The application of economics in animal health programmes: a practical guide. Rev, sci. tech. Off. Int. Epiz. 18, 305-314

Office International des Épizooties (1999). The economics of animal disease control. 18, 265pp

Ott, S.L., Seitzinger and A.H. and Hueston, W.D. (1995) Measuring the national economic benefits of reducing livestock mortality. Prev. Vet. Med. 24, 203-211

Perry, B.D. and Randolph, T.F. (1999). Improving the assessment of the economic impact of parasitic diseases and of their control in production animals. Vet. Para. 84, 145-168

Power, A.P., and Warwick, K.S. (1984). An approach to the measurement of the costs and benefits of poultry meat inspection. Proceedings of the Society for Veterinary Epidemiology and Preventive Medicine, 90-103.

Ramsey, G.C., Philip, P. and Riethmuller, P. (1999). The economic implications of animal diseases and disease control at the national level. Rev, sci. tech. Off. Int. Epiz. 18, 343-356

Rushton, J., Thornton, P.K. and Otte, M.J. (1999). Methods of economic impact assessment. Rev, sci. tech. Off. Int. Epiz. 18, 315-342

Sanjuán, A.I., Dawson, P.J. (2003). Price transmission, BSE and structural breaks in the UK meat sector. Eur. Rev. Agric. Econ. 30, 155-172

Sugden, R. and Williams, A. (1978). The Principles of Practical Cost-Benefit Analysis. Oxford University Press. 275 pp

Tambi, E.N., Maina, O.W., Mukhebi, A.W. and Randolph, T.F. (1999). Economic impact assessment of rinderpest control in Africa. Rev, sci. tech. Off. Int. Epiz. 18, 458-477

Valle, P.S., Skjerve, E., Martin, S.W., Larssen, R.B., Østerås, O. and Nyberg, O (2000). A cost benefit evaluation of the Norwegian bovine virus diarrhoea control and eradication program. Proceedings of the 9th International Symposium on Veterinary Epidemiology and Economics, Breckenbridge, Colorado

COMPARISON OF THE CHARACTERISTICS OF DAIRY AND NON-DAIRY FARMING HOUSEHOLDS IN A RURAL SMALLHOLDER COMMUNITY IN NORTHERN VIETNAM

K. SUZUKI[1], M. KANAMEDA AND D.U. PFEIFFER

SUMMARY

The main objective of this field questionnaire study was to provide information about the characteristics of rural smallholder farming communities which have been targeted by the Vietnamese government's dairy development policy. Particular emphasis was placed on the socio-economic characteristics that differentiate between households adopting or not adopting dairy farming. A total of 99 dairy farming households (11 per commune) and 198 non-dairy farming households (22 per commune) were randomly recruited from 9 of 32 communes in Ba Vi District, Ha Tay Province, Northern Vietnam, using two-stage cluster sampling.

The results from this study suggest that the adoption of dairy farming is partly determined by pre-existing wealth of the farming households. It is therefore likely that while the government's dairy development policy has achieved its first objective of an increase in the dairy cattle population, it has been less successful at achieving the second objective of reducing the level of poverty within rural areas.

INTRODUCTION

Vietnam is a significant exporter of rice, whereas most countries in Southeast Asia are barely able to meet domestic demand (Anon., 2001a). Rice export has heavily contributed towards national economic growth, however, average household incomes are still low and poverty is severe in many areas in Vietnam (World Bank, 1999). Although intensification of rice production will be important, poverty reduction is expected to be achieved primarily through increased diversification of cropping, as well as non-crop activities such as intensive livestock production. Large and small ruminants, pigs and poultry currently are a minor but necessary component of income generation (Dixon et al., 2001).

The Vietnamese government has developed a policy aimed at the introduction of dairy production in order to alleviate poverty amongst rural smallholder communities (Anon., 2000). As a result, the number of dairy cattle has increased in recent years (Do & Hoang, 2001), but further increases will depend upon whether it can be demonstrated that farming households benefit economically from this development. Past experience in various developing countries has shown that such developments can result in unsustainable systems, when they are not based on an understanding of production systems. Field surveys therefore have to be conducted to

---

[1] Kuniaki Suzuki, Epidemiology Division, Department of Veterinary Clinical Sciences, The Royal Veterinary College, Hawkshead Lane, North Mymms, Hatfield, Herts., AL9 7TA, UK. Email: ksuzuki@rvc.ac.uk.

describe the current situation and consider the sustainability of the development (Kaasschieter et al., 1992).

There is relatively little published information on most aspects of dairy production at the smallholder level in Vietnam (Anon., 1995; Do & Hoang, 2001). Several socio-economic surveys have been conducted to describe the complex and diverse integration of various crops, animal species and aquaculture in Vietnam (Devendra et al., 1997). These include a review of smallholder dairy production systems (Anon., 1995) and a survey of animal production amongst 200 smallholder households in a province in the northern midlands (Landon-Lane et al., 1996). The following findings were reported:

- Livestock numbers and species increased with wealth level. The largest increase was in the number of poultry, followed by pigs.
- The value of production from each component enterprise of the farming system increased proportionally with household wealth.
- Livestock production was the second largest component of the farming system after rice, in terms of value at all wealth levels.

Two farming household and animal health baseline surveys were conducted by a European Union Project in support of government veterinary services in Vietnam. One of the surveys was conducted in Ha Tay Province which is the location where Ba Vi District, the present study site, is located (Anon., 2001b). The World Bank conducted two nationwide living standards surveys (World Bank, 1994; 2001) and several reports were published based on this information (World Bank, 1999; Anon., 2003; Justino & Litchfield, 2003). These survey findings need to be interpreted with care, as the sampling methods used were not optimal for the intended purpose.

The objective of the current survey was to provide information about the characteristics of the smallholder farming communities in which the Vietnamese government's dairy development policy has been implemented. Particular emphasis was placed on the socio-economic characteristics that differentiate between households adopting or not adopting dairy farming. This objective is part of a larger investigation into constraints to dairy health and production in smallholder communities in northern Vietnam which includes a longitudinal study amongst the dairy farmers included in this survey.

MATERIALS AND METHODS

Study area

Ba Vi District, which is part of Ha Tay Province in Northern Vietnam, was selected as the study area. It was considered representative of the farming areas in the vicinity of Hanoi. The main agriculture activities in Ba Vi District are the cultivation of rice and to a lesser extent livestock keeping and the production of fruits and tea leaves. The main cattle production system in this area has been based for many years on the Vietnamese Yellow Cattle breed, which was kept for fattening, draught power and manure production purposes. More recently, dairy cattle production has been encouraged by the government to satisfy the demand for milk products in urban areas such as Hanoi.

According to the District Veterinary Station (DVS) in Ba Vi, there are about 500 households involved in dairy cattle farming amongst a total of approximately 52,000 households in the district. The total number of dairy cows is 1,100, which represents about 2.2 cows per dairy cattle farming household (DVS, 2001, personal communication).

The mean annual temperature in the study area is 21.5°C with minimum and maximum averages of 16.4°C in January and 28.9°C in July, respectively. The area receives an average rainfall of 1500-1600mm per year with the highest rainfall during the period June-September (300mm per month) and the lowest rainfall during December-March (less than 40mm per month). Grass growth is year round but is limited during the dry, cool months from October-February (Holland et al., 2000).

Selection of study farms

Primary sampling units were selected using random two-stage cluster sampling amongst the two groups of farms (dairy and non-dairy) where dairy cattle farming had been developed in the Ba Vi district prior to the time of selection (9 out of 32 communes in Ba Vi District). Staff from the DVS provided access to a register of the agricultural households in each commune. As secondary sampling units, 11 dairy and 22 non-dairy households were randomly recruited from the register of each of the 9 communes, resulting in a total of 99 dairy and 198 non-dairy households. The sample size was sufficient to detect odds ratios of 2.25, assuming an exposure prevalence of 20% amongst unaffected households, at a statistical power of 80% and significance level of $p < 0.05$.

Location of study area

Figure 1 shows the location of the selected communes within the study area.

Questionnaire and interviews

A questionnaire was designed to obtain basic information about the two agricultural household groups included in this survey. Major sections comprised of questions relating to household characteristics, holding land area, livestock composition, self-declared cash income as well as future plans in relation to the farming operation. If the interviewers had doubts with respect to the accuracy of the responses, DVS staff were consulted for confirmation after the interview.

Each household's economic wealth was assessed visually by the interviewer based on a set of defined indicators. The condition and structure of the house such as actual age of house (as is the local custom, year of house construction was usually carved on a ceiling beam or painted on the wall above the entrance door), building material (simple-local/ solid-modern), number of rooms and general condition of house were assessed. A variable 'general condition of house' was obtained through an overall evaluation of the farm house and whole compound based on tidiness and cleanliness. In addition, the ownership of selected farm and household items such as motor pump, bicycle, motorbike/car, radio, TV and video recorders was determined.

The questionnaire was translated into Vietnamese by staff of the National Institute of Veterinary Research in Hanoi. The questionnaire was pretested on five households, after which a small number of changes were made to improve the clarity of questions. Questionnaire interviews were conducted by one member of the DVS staff in the presence of the first author along with a commune-based animal health worker, between 26 March and 15 May 2002. The first author performed the visual assessment of each household's economic wealth. The commune-based animal health workers acted as guides. The average duration of interviews of dairy and non-dairy farming households was 40 and 20 minutes, respectively.

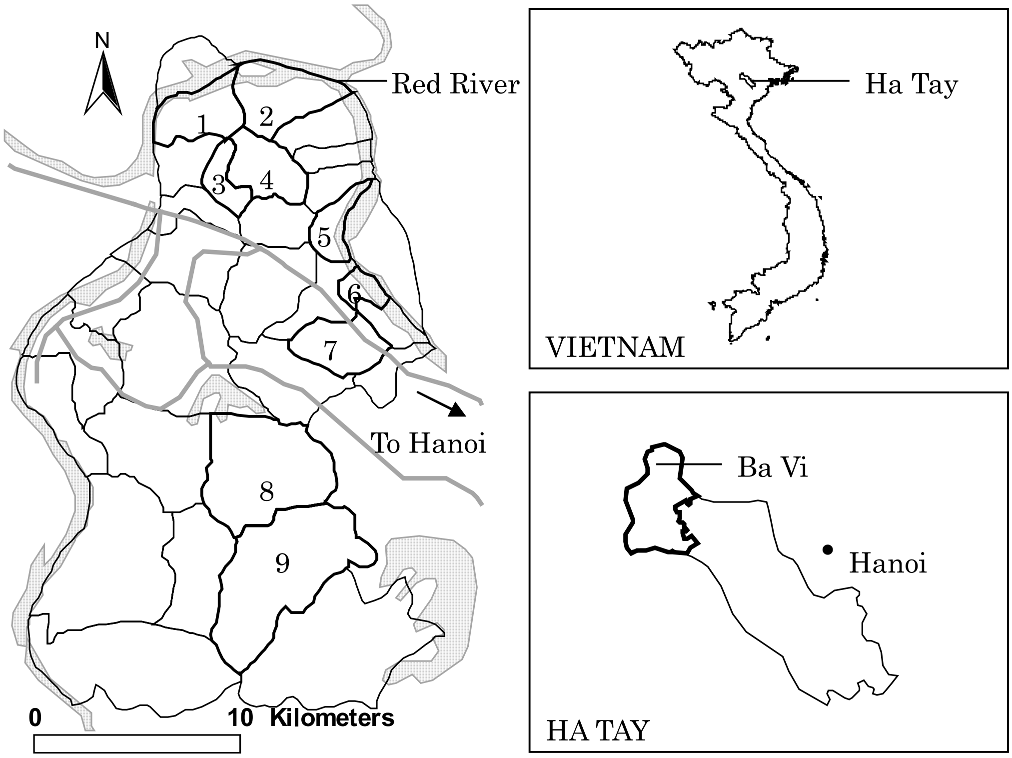


Fig. 1 A map of Ba Vi District, showing the location of the nine survey communes (1 Co Do, 2 Phu Cuong, 3 Phu Dong, 4 Van Thang, 5 Phu Chau, 6 Chu Minh, 7 Tien Phong, 8 Tan Linh and 9 Van Hoa)

Data analysis

Data collected during the two-month period in 2002 was entered into a database in Microsoft Access® 2002 (Microsoft Corporation, Redmond, USA). Maps were produced using the geographical information system software ArcView® for Windows version 3.2 (ESRI Inc., Redlands, USA). The statistical analyses were performed using SPSS® for Windows version 11.5 (SPSS Inc., Chicago, Illinois, USA) and NCSS 2000 (NCSS statistical software, Kaysville, Utah, USA).

Univariable and multivariable analyses were used to firstly describe the differences between the two farming household groups categorised according to involvement in dairy farming ('DAIRY' and 'non-DAIRY' farming households). Univariable analyses were conducted using Pearson's chi-squared statistic for categorical factors and Mann-Whitney U tests with continuous scale variables. Exact tests of significance were used when the assumptions of standard asymptotic statistical methods were not met. Following the univariable analyses, an exploratory multivariable analysis was conducted using multiple correspondence analysis (MCA) to further understand the relationships between categorical variables. In a multivariable logistic regression analysis, the most important factors differentiating between 'DAIRY' and ‘non-DAIRY’ farming households were identified, based on inclusion of all variables, which were significant at $p < 0.10$ in the univariable analysis. A stepwise backward variable selection approach was used based on the likelihood-ratio statistic and entry and removal probabilities of $p < 0.05$ and $P > 0.10$, respectively. The model goodness-of-fit was assessed using the Hosmer-Lemeshow goodness-of-fit test. All variables included in the final regression model were screened for possible interactions and, in case of continuous variables, for non-linearity of effects (Katz, 1999).

RESULTS

Descriptive analysis

The 99 dairy and 198 non-dairy households interviewed accounted for about 20 and 0.4% of the total dairy and non-dairy farming households in the study area, respectively. Amongst the DAIRY farmers, the mean number of years of experience in dairying was 2.7 years (range 1-7 years). Almost 80% of the DAIRY farmers started dairy farming in or after the year 1998 with 85% of them receiving a formal or informal sector loan when purchasing dairy cattle.

Univariable analysis

Household characteristics: On average, the household heads were in their mid-forties and had to look after five household members, including themselves (Table 1). DAIRY farmers (36%) were more likely to have an upper secondary or higher level of education (versus up to lower secondary or compulsory level) compared with non-DAIRY farmers (21%; $\chi^2 = 7.82$, $p < 0.01$). Only three percent of the DAIRY group employed a small number of farm workers (range 2-4 people), whereas the non-DAIRY only employed family members ($\chi^2 = 6.06$, $p = 0.04$).

Table 1. Characteristics of households stratified by farming category.

Farming category	N	Mean age of household heads (range)	Mean size of households (range)	Mean number of adults [> 18 years of age] (range)	Mean number of children (range)
Dairy farming households	99	44.7 (19-78)	5.0 (2-9)	3.2 (2-7)	1.8 (0-5)
Non-dairy farming households	198	46.8 (21-81)	5.1 (2-10)	3.2 (2-7)	1.9 (0-6)
P value		0.45	0.82	0.75	0.63
Mann-Whitney U statistic		9277.5	9641.5	9588.5	9477.0

Land holding area and use: Table 2 shows the comparison of land holding area, classified according to agricultural usage, between the two farming groups. In all of the land use classifications, except for ‘garden’, the land holding area of DAIRY farmers was significantly larger than that of non-DAIRY farmers. DAIRY farmers (35%) were more likely to own a fish pond than non-DAIRY farmers (15%; $\chi^2 = 16.7$, $p < 0.01$, odds ratio 3.2, 95%C.I. = 1.8-5.6).

None of the households owned forest or pasture land. Only two percent of DAIRY leased out a small area of land for cultivation (range 0.04-0.08 hectares; $\chi^2 = 4.03$, $p = 0.11$).

Table 2. Relationship between farming category and land use.

Farming category	Land area owned [Mean (range), hectares] Total	House yard	Cultivated area	Garden[a]	Fish pond[a]
Dairy farming households	0.51 (0.03-2.8)	0.02 (0-0.7)	0.34 (0-2.6)	0.12 (0-1.4)	0.18 (0.01-1.2)
Non-dairy households	0.29 (0.03-1.1)	0.01 (0-0.04)	0.19 (0-1.0)	0.10 (0-0.9)	0.05 (0.01-0.4)
P value	<0.001	<0.001	<0.001	0.21	0.049
Mann-Whitney U statistic	5034.0	7125.5	5134.5	4607.5	362.5

[a]The farming households not holding land in a particular land use category were excluded.

Livestock composition: All households kept a range of livestock species (Table 3). DAIRY farmers were less likely to own pigs or chickens. When households not owning a particular livestock species were excluded, the average number of chickens kept by DAIRY farmers was significantly higher than that of non-DAIRY farmers. Although DAIRY farmers were less likely to own buffaloes and ducks than non-DAIRY farmers, these differences were not statistically significant. The percentage of farmers involved in fish farming completely agreed with the data relating to fish pond ownership, described above.

Self-declared cash income: Annual gross on- and off-farm income was investigated according to different categories of farm products and non-agricultural income sources (Table 4). Compared with non-DAIRY, DAIRY earned significantly more on-farm income, even if the income from dairy farming was excluded. However, DAIRY (22%) was less likely to have one or more sources of off-farm income in addition to on-farm activities, compared with non-DAIRY (55%; $\chi^2 = 28.0$, $p < 0.01$).

There were a wide variety of off-farm income sources such as salary from employment (e.g. factory worker, craftsman, teacher), pension from military service and other businesses (e.g. stall, bicycle repair, furniture making). DAIRY (10%) was less likely to be in salaried employment than non-DAIRY (31%; $\chi^2 = 16.2$, $p < 0.01$). In contrast to non-DAIRY (15%), no DAIRY farmer received a pension ($\chi^2 = 16.7$, $p < 0.01$). There was no difference between DAIRY (12%) and non-DAIRY (9%) in the proportions that owned a business other than farming ($\chi^2 = 0.94$, $p = 0.33$).

Table 3. Relationship between ownership of different livestock species and farming category.

Variable	Dairy farming households	Non-dairy farming households	P value	χ^2 statistic	Mann-Whitney U statistic
Ownership (%)					
Buffaloes	9%	17%	0.06	3.5, 1 df	
Dairy cattle	100%	0%	-	-	
Non-dairy cattle	40%	63%	<0.001	13.8, 1 df	
Pigs	60%	96%	<0.001	64.4, 1 df	
Chickens	26%	71%	<0.001	52.9, 1 df	
Ducks	1%	4%	0.28[b]	2.1, 1 df	
Fish	35%	15%	<0.001	16.7, 1 df	
Mean no. (range)[a]					
Buffaloes	1.4 (1-5)	1.5 (1-3)	0.21		118.0
Dairy cattle	2.4 (1-8)	0	-		-
Non-dairy cattle	1.4 (1-4)	1.3 (1-3)	0.55		2382.5
Pigs	4.9 (1-30)	5.4 (1-33)	0.12		4871.5
Chickens	56 (10-200)	30 (10-100)	0.01		1266.0
Ducks	20	158 (30-400)	0.22[b]		0.0

[a] The farming households not holding livestock species mentioned in each row were excluded.
[b] Exact significance

Table 4. Relationship between farming category and self-declared annual cash income.

Category	Self-declared annual cash income [Mean (SD), million VND[a]]			
	Total income	On-farm income, dairy income excluded	Dairy income	Off-farm income[b]
Dairy farming households	22.2 (15.1)	11.0 (10.1)	8.7 (9.0)	11.3 (9.4)
Non-dairy farming households	10.3 (6.0)	7.8 (5.2)	0	4.5 (3.4)
P value	<0.001	<0.001	-	<0.001
Mann-Whitney U statistic	3388.0	7267.0	-	349.0

[a] VND - Vietnamese Dong, exchange rate USD/VND as of May 2002: 1/15,000
[b] The farming households not receiving off-farm income were excluded.

Future plans in relation to farming operation: The majority of households did not intend to make any changes to their current approach to crop production (87% of DAIRY and 82% of non-DAIRY; χ^2 = 1.01, p = 0.32). A small number of farmers expressed an interest in developing pasture and starting fruit production (4% each amongst DAIRY) and expanding an existing orchard (13% of non-DAIRY). Most DAIRY wanted to increase the number of dairy cattle (79%) and 55% of non-DAIRY intended to begin dairy farming. Some non-DAIRY farmers (26%) wanted to increase the number of pigs. No change with respect to livestock production was planned by 21% and 5% of DAIRY and non-DAIRY farmers, respectively (χ^2 =

18.4, $p < 0.01$). DAIRY (87%) as well as non-DAIRY farmers (92%) considered lack of capital to be their most important problem ($\chi^2 = 2.39$, $p = 0.12$). Amongst DAIRY farmers, livestock (71%) was the most important source of income, followed by off-farm activities (13%), rice (8%) and other on-farm activities (8%). Important sources of income for non-DAIRY farmers were rice (39%), followed by livestock (36%), off-farm activities (20%) and other on-farm activities (5%; $\chi^2 = 41.9$, $p < 0.01$).

Indicators of economic wealth: There was no difference in the average age of the house between DAIRY and non-DAIRY households (median 13 and 14 years, range <1-102 and 1-102 years, respectively, $p = 0.20$). DAIRY households were more likely to own a 'modern' house than non- DAIRY households (solid-modern vs. simple-local; 97% and 87%, respectively; $p = 0.01$), have more rooms in the house (4 or over vs. 1 to 3; 14 and 6%, respectively; $p = 0.01$) and have improved general condition (clean-tidy vs. other; 71 and 51%, respectively; $p < 0.01$). All households had electricity supply. As there was no permanent water supply infrastructure in the study area, households either had a well or a rainwater tank on their properties. DAIRY was more likely than non-DAIRY to own the following items: motor pump (90 and 56%, respectively; $p < 0.01$), motorbike/ car (57 and 39%, respectively; $p = 0.01$), TV (93 and 75%, respectively; $p = 0.01$) and video recorder (36 and 24%, respectively; $p = 0.03$). Almost all non-DAIRY households (99%) owned a bicycle, whereas less DAIRY households did own one (86%; $p < 0.01$). There was no significant difference in the proportion of radio ownership between DAIRY (42%) and non-DAIRY households (45%).

Multivariable analysis

MCA of economic wealth indicators: Figure 2 displays the differences between the two farming groups with regard to the structure and condition of the house and the existence of selected farm and household items. The plot explains 57.1% of the total inertia using two dimensions. The first dimension focuses more on the farm and household items, and the second dimension on the characteristics of their house. Separation in the supplementary variable DAIRY or non-DAIRY occurs mainly in relation to the first dimension, and to a limited extent along the second dimension. It shows that DAIRY households were more likely to own a tidy, large, modern house and a range of more expensive items such as a motor pump, motorbike/car, TV and video recorder than non-DAIRY households.

Multiple logistic regression: The final multiple logistic regression model included variables relating to livestock species kept by the household, income sources and economic wealth indicators (Table 5). The variables relating to salaried employment or receiving a pension were combined into a single variable to prevent zero frequencies in the variable 'receiving a pension' from affecting effect estimation. The resulting new variable 'receiving a salary or pension' represented the off-farm income category other than 'owning a business'. The final model indicates that DAIRY farmers were less likely to own buffaloes, non-dairy cattle, pigs and chickens compared with non-DAIRY farmers. Also they were two and a half times as likely to be involved in fish farming as non-DAIRY farmers. DAIRY farmers were more likely to have income from a diverse range of on-farm and off-farm sources other than traditional rice cultivation. They were less probable to be in salaried employment or receive a pension than non-DAIRY farmers. Living in a better house, owning a motor pump and not owning a bicycle were more likely to be characteristics of DAIRY farmers than non-DAIRY farmers.

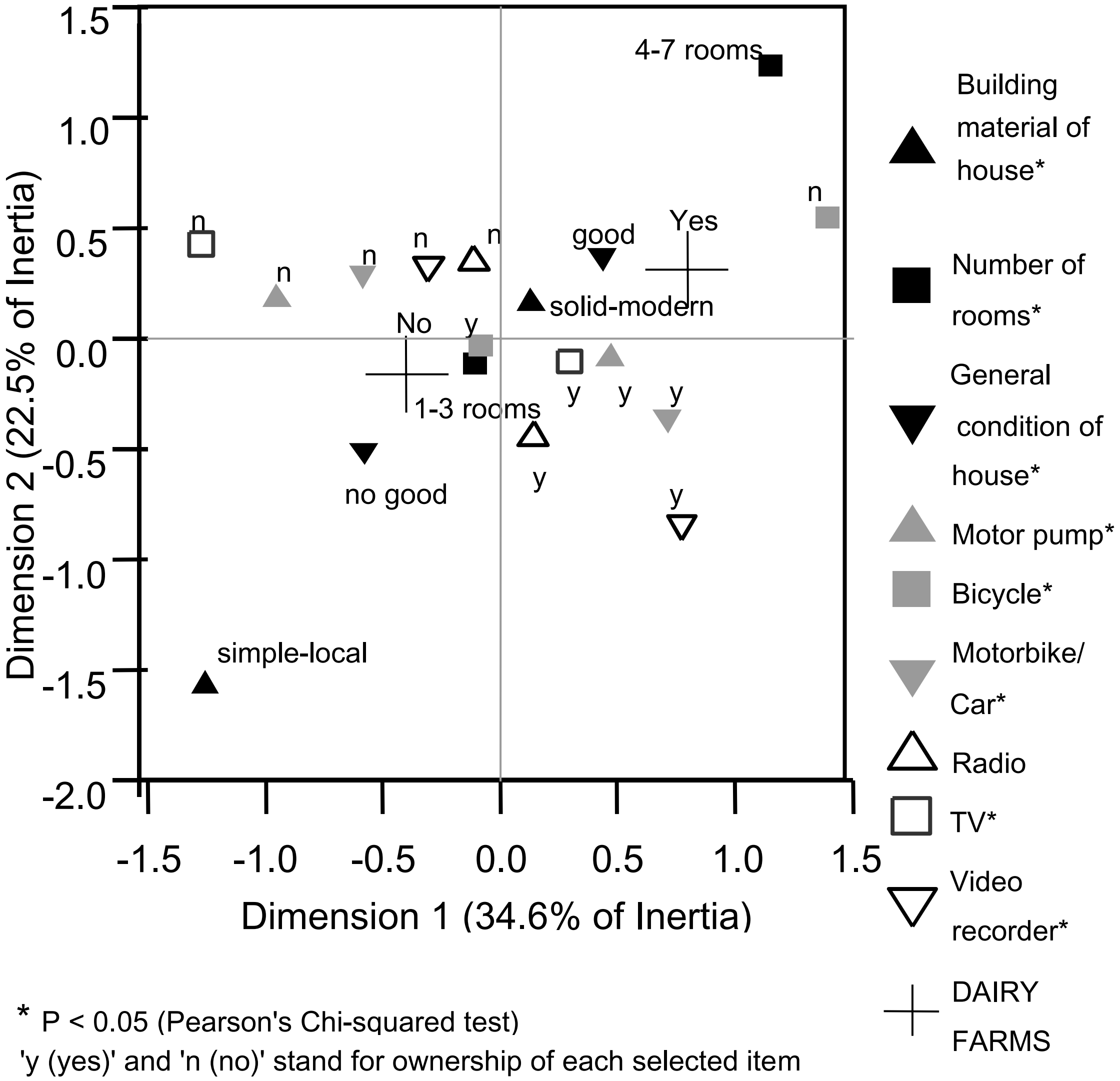


Fig. 2 2D plot based on MCA of indicators of farmer's economic wealth. The indicators of farmer's economic wealth are displayed as column coordinates (Col) while the farming category is added as supplementary column points (+)

DISCUSSION

The difficulties associated with conducting field surveys for animal health and productivity issues in developing countries such as Vietnam have been described previously (Thrusfield, 1995) and, one of the main pre-requisites are the co-operation and goodwill of farmers (Cameron, 1999). In Vietnam, a number of surveys have been carried out by donor agencies, but most have failed, as a result of communication problems between donor agencies and farmers (Anon., 2001b; Perkins, 2002). When each household was visited during the current study, the interviewers carefully developed a good relationship with farmers and explained the importance of the survey. As a result, the survey achieved 100% participation amongst the farmers selected and no responses to individual questions were refused.

The results from this study suggest that the adoption of dairy farming is partly determined by pre-existing wealth of the farming households. Holding a larger land area, obtaining more on-farm income (excluding dairy income), owning a better quality house and more selected farm and household items were interpreted as indicators of pre-existing wealth amongst households belonging to the DAIRY group. The mean number of years of experience in dairying was relatively short (2.7 years) and 85% of them received a loan for purchasing dairy cattle. Generally, farmers in Ba Vi District started dairy farming with an in-calf heifer costing 25-28 million Vietnamese Dong (Nguyen, 2001, unpublished data). This is higher than the average self-declared annual cash income of the DAIRY group (22 million Vietnamese Dong). This suggests that it would not be feasible for the farmers to use the profits made from dairy farming up until the time of the questionnaire survey for improving the economic wealth, as reflected by the indicators measured in this study. Instead, most dairy farmers in the study area will have to pay off their loan over a number of years.

Table 5. Final multiple logistic regression model describing the probability of a household having a DAIRY farming operation in relation to various household characteristics.

Independent variable	P value	Odds ratio	95% CI Lower	95% CI Upper
Ownership of livestock species				
Buffaloes	0.039	0.32	0.11	0.95
Pigs	<0.001	0.03	0.01	0.16
Chickens	<0.001	0.17	0.08	0.36
Ducks	0.021	0.06	0.01	0.65
Fish	0.036	2.54	1.06	6.10
Most important income source				
Rice (reference)				
Livestock	<0.001	11.69	3.30	41.37
Other on-farm income	0.026	7.26	1.26	41.76
Off-farm income	0.039	4.89	1.08	22.13
Receive a salary or pension	0.002	0.18	0.06	0.54
House built with solid/ modern materials	0.057	3.97	0.96	16.43
Ownership of farming and household items				
Motor pump	0.009	4.28	1.44	12.73
Bicycle	0.017	0.11	0.02	0.67

Hosmer-Lemeshow goodness-of-fit statistic 2.6, 8 df, p = 0.96

The data presented in Table 5 shows that dairy farmers are less likely to own livestock species other than dairy cattle than non-dairy farmers. This suggests that DAIRY farmers have focused their livestock-related activities towards dairy cattle, as dairy farming is considered quite labour-intensive (Nguyen, 2001, unpublished data). This interpretation is supported by the fact that there was no difference in the average household size between the two farming groups (Table 1). In other words, DAIRY farmers reduced diversification rather than employing additional staff from outside of the household members. Fish farming requires access to sufficient land area for setting up a pond, which is easier for DAIRY farmers as they own larger land areas (Table 2).

The majority of households did not plan any changes with respect to crop production in the immediate future. One possible explanation for this position in the case of dairy farmers may be that they are satisfied with the profitability of their dairy farming operation and therefore prefer to maintain the same level of crop production. Amongst non-DAIRY farmers, crop production does not satisfy their subsistence requirements, and since many of them own a limited area of land, there is no actual potential for increasing crop production (Table 2). As a result, most non-DAIRY farmers engage in additional off-farm activities (Table 4) to satisfy their subsistence requirements. In rural Vietnam, the World Bank (1999) estimated that the share of household income derived from off-farm activities accounted for 53%.

In relation to possible changes in livestock production, both DAIRY and non-DAIRY farmers indicated a high level of interest in dairy farming. Most DAIRY farmers did not want to make any changes to livestock production other than increasing the number of dairy cattle. This shows the degree of satisfaction with this component of their farming operation, presumably mainly as a result of the economic benefits received. Almost three-quarters of DAIRY farmers considered livestock (mainly dairy cattle) to be their most important income source. The analysis in relation to farming operation indicates that dairy farming represents a higher economic value for DAIRY. Non-DAIRY farmers selected rice followed by livestock (almost all of which was pig farming) as their most important income source. This is in agreement with the estimates of the share of agricultural revenue in farming households in rural Vietnam reported by the World Bank (1999) which ranked rice first (44%) and livestock (31%).

DAIRY farmers were more likely to own farm and household items, such as motor pumps, motorbikes or cars, TVs and video recorders, than non-dairy farmers (Figure 2). When dairy farming was first introduced to smallholders in the study area, governmental extension officers recommended purchase of motor pumps to farmers for daily management of the cows such as supply of water and for cleaning purposes (Nguyen, 2001, unpublished data). Oddly, bicycles were less likely to be owned by DAIRY (86%) compared with non-DAIRY farmers (99%). The World Bank (1999) has reported that rural people in Vietnam without access to motorised transport services were nearly five times more likely to be found in the poorest quintile than in the richest quintile. In the current study, wealthier DAIRY farmers would have replaced bicycles with motorbikes as a means for transportation.

It is interesting that DAIRY households were also more likely to have a diverse range of income sources (other than traditional rice cultivation) than the non-DAIRY group (Table 5). Unfortunately, the study design will not allow conclusions with respect to whether diversification was the result of pre-existing wealth or if it was a strategy that was more likely to lead to increased wealth.

CONCLUSION

The results of the field questionnaire survey show that there are considerable differences between dairy and non-dairy farms in relation to household wealth. This suggests that the adoption of dairy farming is partly determined by pre-existing wealth of the farming households. At present, it is unclear whether the dairy farming development can result in poverty alleviation in the study area. The effect of the Vietnamese government's dairy development policy appears to be that while the dairy cattle population has increased it is unlikely to impact on the existing disparity in wealth within rural areas.

ACKNOWLEDGEMENTS

The study was carried out as part of the Vietnam-Japan project ‘Strengthening Capacity of the National Institute of Veterinary Research’ at the National Institute of Veterinary Research, Hanoi, Vietnam. The authors wish to thank the farmers who allowed us to interview them in relation to their farming and other activities, and the staff of the National Institute of Veterinary Research and the District Veterinary Station in Ba Vi for supporting us during the implementation of the field questionnaire survey. The study was funded in part by the Japan International Co-operation Agency.

REFERENCES

Anon. (1995). Proceedings of the workshop on improved feeding systems for dairy cattle and milk production in smallholder farms. Institute of Agricultural Science of South Vietnam, Ho Chi Minh City, Vietnam. 141p

Anon. (2000). Project for development of dairy production in Vietnam 2000 to 2010. Ministry of Agriculture and Rural Development (MARD), Hanoi, Vietnam.

Anon. (2001a). Statistical yearbook 2000. General Statistical Office. Statistical Publishing House, Hanoi, Vietnam. 599p

Anon. (2001b). Strengthening of Veterinary Services in Vietnam (SVSV) Project Agro-economic farm household & animal health baseline survey: Phu Cat/Tan Phu Communes [7-8/2000] - Quoc Oai District, Ha Tay Province. MARD and the European Commission Project for SVSV, Hanoi, Vietnam.

Anon. (2003). Income diversification and poverty in the northern uplands of Vietnam. International Food Policy Research Institute (IFPRI) and Japan Bank for International Cooperation (JBIC). Japan Bank for International Cooperation, Tokyo, Japan. http://www.jbic.go.jp/english/oec/environ/report/vietnam.php

Cameron, A.R. (1999). Survey toolbox - A practical manual and software package for active surveillance of livestock diseases in developing countries. Australian Centre for International Agricultural Research (ACIAR), Canberra, Australia. 330p

Devendra, C., Thomas, D., Jabbar, M.A. and Kudo, H. (1997). Improvement of livestock production in crop-animal systems in rainfed agro-ecological zones of South-east Asia. International Livestock Research Institute, Nairobi, Kenya. 116p

Dixon, J., Gulliver, A. and Gibbon, D. (2001). Farming systems and poverty. Improving farmers' livelihoods in a changing world. Food and Agriculture Organisation, Rome, Italy. 413p

Do, K.T. and Hoang, K.G. (2001). Dairy cattle production in Vietnam and development plan for 2002-2010. Ministry of Agriculture and Rural Development, Hanoi, Vietnam.

Holland, W.G., Luong, T.T., Nguyen, L.A., Do, T.T. and Vercruysse, J. (2000). The epidemiology of nematode and fluke infections in cattle in the Red River Delta in Vietnam. Vet. Parasitol. 93, 141-147

Justino, P. and Litchfield, J. (2003). Welfare in Vietnam during the 1990s: Poverty, inequality and poverty dynamics. PRUS Working Paper No. 8. Poverty Research Unit at Sussex, University of Sussex, Brighton. http://www.sussex.ac.uk/Units/PRU/wps/wp8.pdf

Kaasschieter, G.A., Dejong, R., Schiere, J.B. and Zwart, D. (1992). Towards a sustainable livestock production in developing countries and the importance of animal health strategy therein. Vet. Q. 14, 66-75

Katz, M.H. (1999). Multivariable analysis: A practical guide for clinicians. Cambridge University Press, Cambridge, UK. 192p

Landon-Lane, C., Kinh, N.N., Hung, N.D. and Ban, B.K. (1996). Customers for animal science research in Vietnam: What do they need? In: Pryor, W.J. (Ed.) Exploring approaches to research in the animal sciences in Vietnam. ACIAR, Canberra, Australia. pp. 190-194

Perkins, J. (2002). Profitable beef cattle development in Vietnam: Project review and final workshop-Review mission briefing papers. ACIAR, Hanoi, Vietnam. 62p

Thrusfield, M.V. (1995). Veterinary epidemiology. 2nd edn. Blackwell Science Ltd, Oxford, UK. 504p

World Bank (1994). Viet Nam living standards survey (VNLSS), 1992-93 basic information. Washington D.C., USA. http://www.worldbank.org/lsms/country/vn93/vn93docs.html

World Bank (1999). Vietnam development report 2000: Attacking poverty. Joint report of the government, donor and NGO working group. Consultative Group Meeting for Vietnam, Hanoi, Vietnam. http://www.vdic.org.vn/eng/pdf/attpov.pdf

World Bank (2001). Viet Nam living standards survey (VNLSS), 1997-98 basic information. Washington D.C., USA. http://www.worldbank.org/lsms/country/vn98/vn98docs.html

COST-BENEFIT COST-BENEFIT ANALYSIS OF MASTITIS CONTROL BY SIMULATION: MODEL DESCRIPTION AND PROFITABILITY OF STRATEGIES TARGETING SUBCLINICALLY INFECTED COWS

H. SEEGERS*, C. FOURICHON, D. BILLON AND P. HORTET

SUMMARY

This study aimed to assess the economic effectiveness of different strategies for the control of subclinical udder infections. Assessment was performed using ECOMAST, a dynamic, stochastic simulation model. Control plans combining lactational treatment of subclinically infected young cows, application of a strict culling policy to persistently infected old cows and/or improvement in prevention were evaluated in four different initial udder-health situations. The plans were ranked according to their discounted gross margin. Control plans that incorporated lactational treatment were amongst the most profitable measures when the udder-health situation of the herd was poor. A strict culling policy resulted in a rapid improvement of the bulk milk somatic cell count but it was very often not profitable because of non-fulfilment of the milk quota. Economic implications of country-specific penalties for somatic cell counts and milk quotas should be considered to help in generalizing the results.

INTRODUCTION

Udder health control strategies can be defined as combinations of several elementary preventive and curative actions. To choose a relevant combination of such actions, farmers need an ex-ante assessment of the expected economic worth of the combined actions for their specific herd. Economic worth of a strategy depends on changes induced in expenditures and economic losses, that is, a cost-benefit marginal variation (McInerney et al., 1992). Simulation models can be developed for such an assessment (Seegers et al., 2003). High prevalence of subclinical mastitis and its obvious economic consequence, such as milk price penalties, are still a common problem on many farms. Control of subclinical udder infections relies mainly upon milking hygiene, post-milking teat disinfection and dry-cow therapy. The effectiveness of these actions can be enhanced by culling of persistently infected cows and by antibiotic treatment of infected lactating cows.

The objectives of this paper are to describe the ECOMAST simulation model and to compare the economic worth of different strategies including lactational treatment (LT) and culling of subclinically infected cows implemented according to various applications of these control measures.

---

*Henri Seegers, Veterinary School & INRA, BP 40706, 44307 Nantes Cedex 3, France
Email: seegers@vet-nantes.fr

MATERIALS AND METHODS

Description of the simulation model

ECOMAST (Hortet, 1999; Seegers et al., 2000) is a dynamic time-step model which uses time steps of one day. It is a mechanistic model simulating the herd dynamics indirectly through simulation of each cow within the herd. The herd is represented as a group of cows and heifers, and events are simulated at the cow level. Management decisions are defined at the herd level. It is a stochastic model with the occurrence of discrete events being triggered stochastically which are based on probabilities. Consequently, different runs of the model will result in variable outcomes which will provide a distribution describing the variability of expected results. To run simulation experiments, the same initial herd is simulated over several months or years, firstly where nothing is changed (reference scenario) and secondly, where an alternative control plan is implemented

State variables: Each cow is described by a set of state variables whose values change throughout the simulation. These variables refer to age, reproduction (oestrus, insemination, gestation), milk production (lactation stage, yield, fat, protein), general health and mastitis status of the cow (susceptibility to mastitis, somatic cell count, udder health status), ongoing treatments or milk withdrawal time. Herd-level state variables are also defined to accumulate the individual simulated events and performances. At the beginning of the simulation, state variables are set to values describing the herd of concern.

Driving variables: Driving variables are simple variables or more complex parameterised decision rules. They allow the user to manipulate the simulation model in order to pilot the simulation experiments and to mimic the herd specific management. Values of the driving variables must be defined before starting a simulation. The first group of driving variables is used to define herd and farm parameters (e.g. milk quota, herd size, feeding programme). The second group includes variables used to define decision rules for management of the herd (reproductive strategy, replacement-culling strategy, milk production strategy, quota management and mastitis control strategy). The third group of driving variables are parameters used for economic analysis (e.g. milk pricing system, meat price, cost of each available mastitis control action). The fourth group of driving variables are user-defined parameters which are used in the stochastic simulation processes (e.g. conception rate, probabilities of occurrence of mastitis, probability of evolution of somatic cell count, probability of success of treatments).

General simulation processes: Regarding reproduction, cycling ovarian activity of heifers starts at a user-defined age. Cycling ovarian activity of cows starts after a minimum interval after calving. Heat occurs according to the distribution of interval between calving and first heat or the interval between heats of each animal. Heat is detected according to a detection rate. Decision to inseminate is based on reproductive strategy variables. Result of insemination depends on conception rate. Abortion may occur according to abortion rate. At calving, survival and sex of the calf depend on mortality rate at birth and sex ratio, respectively. Heat, heat detection, conception, abortion, day of calving, calf survival and sex of calf are triggered stochastically.

Daily milk, protein and fat yield are modelled by a 'Wood' equation (Wood, 1967). The result of the 'Wood' equation, which is possibly limited by the feeding plan, is expected yield. Effect of mastitis (and possibly other health disorders) is then subtracted to calculate actual yield. Production level and persistency of each cow are corrected by a lactation number effect to define the 'Wood' equation parameters. Production level and persistency of heifers entering the herd are defined stochastically. Production level of a heifer depends on the production level of her mother and the progress in production level between generations. Feed consumption is deduced from a declared feeding plan and the milk yield of the cows.

Each month, total expected campaign-production is calculated from the current status of the herd as defined by milk yield from the beginning of the campaign and the expected milk yield to the end of the campaign from lactating cows, pregnant cows and heifers. The difference between expected production and quota is calculated. If the difference is within thresholds defined by the user, no specific decision regarding quota management is implemented. If the difference shows an over-production, decisions are made on drying off, culling or selling of cows or heifers. If the difference shows an under-production, decisions are made on purchase of cows or pregnant heifers.

Death and involuntary culling that cannot be delayed are triggered stochastically. Heifers kept in the herd depend on a general strategy (to keep all heifers or to keep a number of heifers sufficient to meet the required replacement rate). Pregnant cows are ranked according to their production level. Heifers born from the 12% cows with the highest or lowest production level are systematically kept or sold, respectively. Culling decisions may occur every 15 days. The need for culling of cows depends on both herd size limits, accumulated milk sales compared to the quota and forecasted calving and drying-off events. If culling is required, all cows are ranked according to a candidacy score. This score combines various criteria (lactation number, reproductive status, milk yield, mastitis and health status). Final culling decisions are made by random selection of the required number of cows within different scoring levels, so as to integrate the imperfect knowledge that the farmer has about the value of each cow and also some non-simulated decision components.

Mastitis simulation processes: Four types of mastitis can be defined, assuming four different categories of pathogens. Definition relies on a set of parameterised characteristics (proportion of clinical and subclinical cases at occurrence of infection, persistence as indicated by rates for self-cure during lactation and at re-calving, effects on yield, milk somatic cell count [SCC], culling or death). The variable risk of occurrence per infection type allows different herd situations to be presented (nature and frequency of infection). Expression and cure rates of the four types of infections were modelled as shown in Table 1.

Table 1. Expression and persistence of the modelled udder-infection types.

Type	% infection with only subclinical expression	Self-cure rate (%) of infection in the current lactation	Cure rate (%) after LT: high efficiency [a]	Cure rate (%) after LT: low efficiency [a]
Coagulase +ve *Staphylococci*	75	20	60	40
Streptococci	40	30	65	55
Gram negative pathogens	10	80	85	85
Minor pathogens	85	50	95	90

[a] Cure rate of 2nd lactation cows after lactational treatment (LT) of subclinical infection

The SCC for a cow at a given day is modelled as the sum of a baseline value without udder infection (function of parity and lactation stage), an additional term according to infection status (type of mastitis, stage in evolution, treatment implemented) and a random variation term. The infection-related additional term is stochastically triggered from a set of possible evolution profiles after onset of infection (declining or flat slope). Reduction in milk yield is modelled from individual cow SCC, as described by Hortet (1999). Every day, each cow is at risk of experiencing each type of mastitis according to a probability defined by a risk function. For each type of mastitis, a baseline cow-level risk function is defined in five different periods with regard to lactation stage. This baseline risk is increased or decreased according to herd-level modifiers (season, prevalence of contagious pathogens and herd-level prevention) and cow-level modifiers (production level, parity, disease history, inter-individual variability and individual prevention).

Herd and initial udder-health situations

This paper reports results for a herd of about 100 Holstein cows producing 8,250 litres each with total solids averaging 43.5 g/l for fat and 33.0 g/l for protein. Usual mastitis control actions are assumed to be intermediate in efficiency (wet udder preparation, postmilking teat disinfection, systematic treatment at drying-off and culling). Culling is mainly based on production and reproductive performance but also takes the individual SCC into account where possible (particularly in managing the quota fulfilment). Purchase of cows or heifers is not allowed and the quota (800,000 l) is always fulfilled despite the prevalence of mastitis and also some cows' milk being used to feed to calves. This last point provides a very demanding context for the model to show any evidence of profitability from the use of additional or modified mastitis control options.

Four initial udder-health situations were studied:

(1) Very high penalties (430,000 cells/ml, 55 clinical cases per 100 cow-years and 1.33 cent[1] /litre milk quality penalties);
(2) High penalties (360,000 cells/ml, 45 clinical cases per 100 cow-years and 1.00 cent /litre milk quality penalties);
(3) Moderate penalties (300,000 cells/ml, 35 clinical cases per 100 cow-years and 0.66 cent /litre milk quality penalties);
(4) Low penalties (260,000 cells/ml, 28 clinical cases per 100 cow-years and 0.33 cent /litre milk quality penalties).

To simulate the predominance of udder pathogens responsible for persistent infections, infection by coagulase-positive *Staphylococci*, *Streptococci*, Gram-negative pathogens and minor pathogens were responsible for 27%, 13%, 5% and 55% of the cases, respectively.

Simulated control plans and utility criterion

Several types of control plans were defined with combinations of LT for young cows, strict culling rules for persistently infected older cows (or, when possible, anticipated drying-off) and improved pre- and post-milking prevention (Table 2). Plans are activated for at least 100 days when bulk-tank milk SCC (BMSCC) is >250,000 cells/ml for 2 months. Lactational treatment and strict culling rules are deactivated when BMSCC decreases to <275,000 cells/ml and

---

[1] Euro cent

improved prevention is deactivated when BMSCC is <100,000 cells/ml. Simulations for simplified plans (including only one action of the combined options) were also run.

Two different costs of LT were simulated, both based on a two-day treatment duration with a four-day withdrawal period (a high cost option of € 27.00 and a low cost option of €22.50). Efficiency was expressed by cure rates (Table 1). Values of cure rates were set lower for older cows and higher for 1st lactation cows.

A 3-year simulation horizon was considered and 100 replications were run for each plan in each situation. The simulation model provided (for each simulated year in a replication) a gross margin (including herd-assets value modification). An annual discount rate of 5% was applied before summing the three annual margins of a replication. Plans resulting in the highest average discounted gross margin, in comparison to the reference scenario, were considered as the most effective. An analysis of variance was used to test the differences between plans.

Table 2. Actions combined in the control plans under study.

Control plan	Code	LT of subclinical mastitis	Culling rules for cows persistently infected	Improved prevention
LT of young cows	LT	LT-2 [a]	No	No
LT extended		LT-3 [b]	No	No
Strict culling above 2nd lactation	Cull	No	C>2 [c]	No
Strict culling above 3rd lactation		No	C>3 [d]	No
Strict culling above 2nd lactation and improved prevention	Cull+Prev	No	C>2 [c]	Yes
Strict culling above 3rd lactation and improved prevention		No	C>3 [d]	Yes
LT of young cows & strict culling	LT+Cull	LT-2 [a]	C>2 [c]	No
LT extended & strict culling		LT-3 [b]	C>3 [d]	No
LT of young cows and improved prevention	LT+Prev	LT-2 [a]	No	Yes
LT extended and improved prevention		LT-3	No	Yes
LT of young cows, strict culling and improved prevention	LT+Cull+ Prev	LT-2 [a]	C>2 [c]	Yes
LT extended, strict culling and improved prevention		LT-3 [b]	C>3 [d]	Yes
Improved prevention	Prev	No	No	Yes

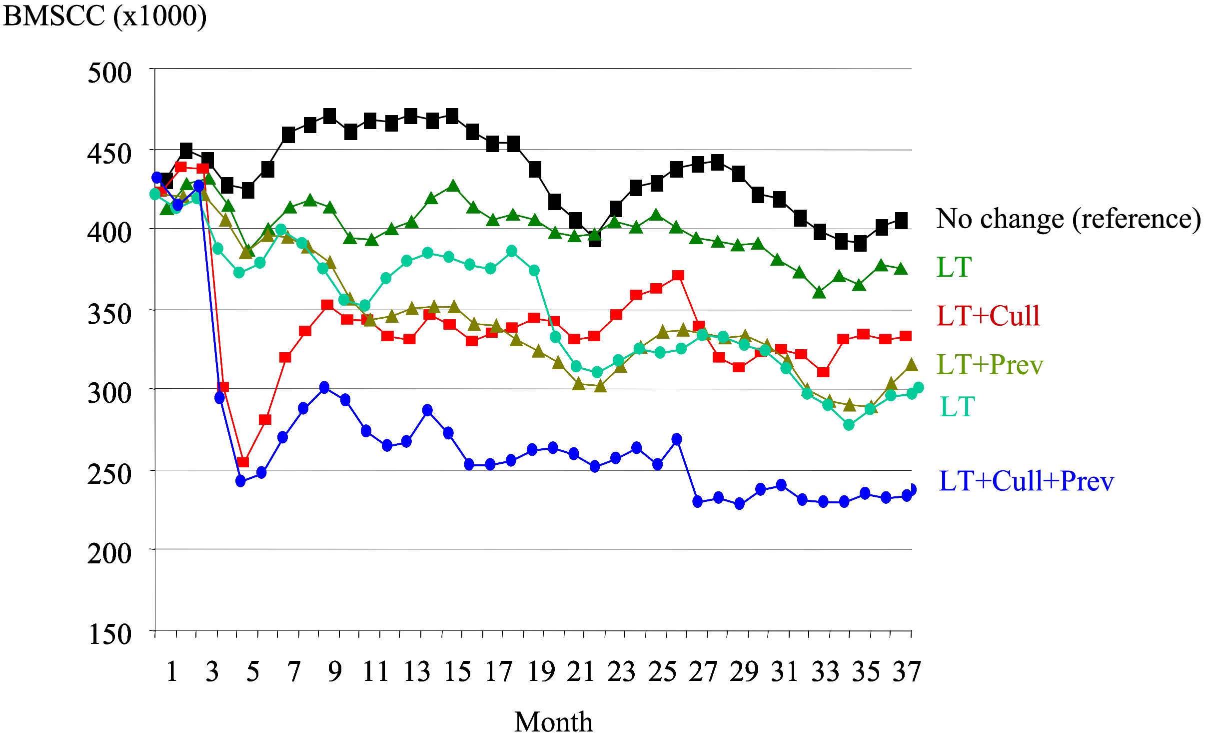
[a] LT of cows in lactation 1 or 2 with two consecutive SCC>300,000 cells/ml
[b] LT of cows in lactation 1 to 3 with one SCC>300,000 cells/ml
[c] Culling (C) of cows in lactation >2 with two consecutive SCC>500,000 cells/ml
[d] Culling (C) of cows in lactation >3 with two consecutive SCC>500,000 cells/ml

RESULTS AND DISCUSSION

Results for each control plan were compared to those of the reference scenario (no change). As an example, Fig. 1 displays some of the average simulated evolution in BMSCC for several control plans starting in the 'very high penalties' initial udder health situation. As shown in Fig.

2, the economic repercussions of the implementation of some control plans were found to be negative, that is, lower than in the reference (no change) scenario.

Fig. 1 Example of the average (100 replications) simulated evolution in BMSCC for some



control plans starting in the ‘very high penalties’ initial udder-health situation, with the assumption of high efficiency of LT.

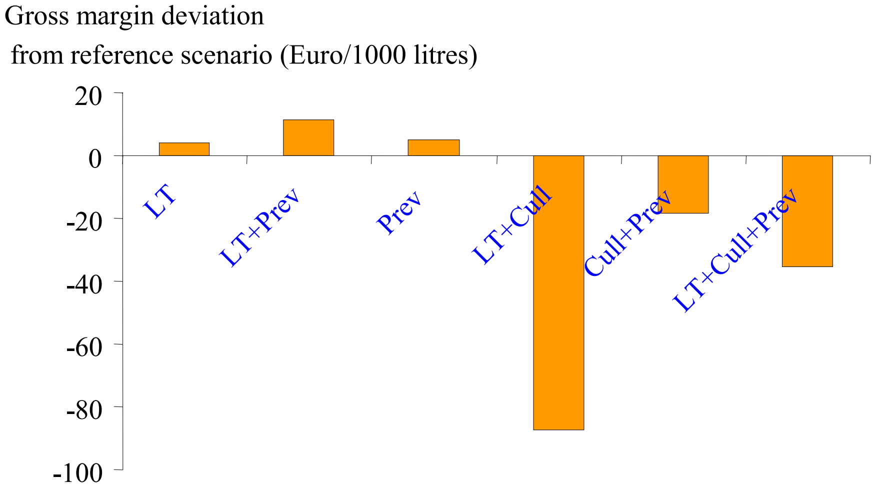


Fig. 2 Example of the average (100 replications) simulated gross margin deviation from the reference scenario (€ /year /1000 litres of quota) for some control plans starting in the ‘very high penalties’ initial udder-health situation, with the assumption of high efficiency of LT treatment. Designation of control plans is referenced to Table 2.

Summary economic results are given in Table 3. The average improvement in gross margin generated by the best plans remained limited reaching only a 3% increase in the best cases. In most of the insufficient udder-health situations considered here, control plans that included LT (according to specific implementation rules) were found to be amongst the most profitable plans. However, LT alone was never found to be a profitable control option. Results varied widely between replications within a same control plan.

Table 3. Gross margin simulation results on the effect of control plans in different udder-health situations compared to the reference plan (no change), assuming a high efficiency of LT.

Initial udder-health situation	Two best control plans in terms of discounted gross margin over a 3-year horizon	Average improvement in gross margin (€ / year / 1000 l of quota)	P
Very high penalties	LT extended and improved prevention	+ 5.7	<0.05
	LT of young cows and improved prevention	+ 3.5	<0.05
High penalties	LT of young cows and improved prevention	+ 6.1	<0.05
	LT extended and improved prevention	+ 4.0	<0.05
Moderate penalties	LT of young cows and improved prevention	+ 3.9	<0.10
	Improved prevention and strict culling	+ 2.2	<0.10
Low penalties	No significant difference with the reference plan	No improvement	NS

The main mechanisms influencing profitability were:

(1) The fulfilment of quota, which is dependent upon the farm culling policy and on the number of target cows for culling. Culling a large number of cows according to systematic rules led to an insufficient number of milk producing cows being kept in herd (in the simulations no external cow purchase was allowed). Strict systematic culling rules did not handicap the quota fulfilment when the number of target cows was low;

(2) The speed of the decline in BMSCC and the associated decrease in milk quality penalties, especially in the 1st year;

(3) The variations in herd-assets value (due to differences in herd size);

(4) The level of extra expenditure required for improved mastitis prevention, which were incurred during the three-year period in most of the udder-health situations that were simulated. The simulated differences in efficiency and cost of LT had only very limited effects (detailed results not shown).

Extrapolation to smaller herds should only be done with caution because the variability of results becomes higher (data not shown). Direct extrapolation to other countries is not valid because the economic contexts are different, particularly in relation to quota management and milk pricing system. Nevertheless, LT could be more profitable than under the French context, where BMSCC penalties are rather low and quotas have strict limits, since there is no quota borrowing system in France and purchase of additional quota is only possible by purchase of farmland that already has quota.

Finally, regarding consumer and society's requests, it is noticeable that LT only led to a short-term increase of the total number of antibiotic treatments in the herd at the beginning of the plan. Later on, the number of subclinically infected cows treated during lactation this was low and was counterbalanced by a lower number of treatments for clinical cases.

CONCLUSION

Within the current French system and in large-sized Holstein herds, antibiotic LT of young cows that are subclinically infected can be advised as a component of mastitis control plans when prevalence of subclinical infections is high and where milk quality penalties represent more than 1.5 % of the price of the milk. Nevertheless, antibiotic LT needs to be combined with improved mastitis prevention in order to reduce the occurrence of new infections. From the economic point of view, to fulfil the quota is the main objective and therefore, culling has to be used with caution as a control action. However, culling is a very efficient process in quickly reducing the BMSCC.

REFERENCES

Hortet, P. (1999). *Ex-ante* assessment of the economic efficiency of mastitis control plans in dairy herds [In French]. Ph.D. thesis, Rennes, France. 148p

McInerney, J.P., Howe, K.S. and Schepers, J.A. (1992). A framework for the economic analysis of disease in farm livestock. Prev. Vet. Med. 13, 137-154

Seegers, H., Fourichon, C., Sørensen, J.T., Hortet, P. and Billon, D. (2000). Modelling the cost-benefits of prevention and control of mastitis in dairy herds. Symposium on Economic Modelling of Animal Health and Farm Management, Wageningen University, Wageningen, The Netherlands, 23-24 November 2000. pp. 55-61

Seegers, H., Fourichon, C. and Beaudeau, F. (2003). Production effects related to mastitis and mastitis economics in dairy cattle herds. Vet. Res. 34, 475-91

Wood, P.H.P. (1967). Algebraic model of the lactation curve in cattle. Nature 216, 164-165.

ANIMAL MOVEMENT

NETWORK ANALYSIS OF THE DANISH CATTLE AND SWINE INDUSTRY TRADE PATTERNS AS AN EVALUATION OF RISK POTENTIAL FOR DISEASE SPREAD: THE HETEROGENEITY ISSUE

M. BIGRAS-POULIN[1], A. THOMPSON, M. CHRIEL, S. MORTENSEN AND M. GREINER

SUMMARY

Trade patterns for animal movements of a specific industry are complex to study because they include many stakeholders, farming sites that are spread heterogeneously over the country, and the flow of animals between the farming sites is very dynamic. Two aspects of this complex situation are of special interest for predicting the outcome of an epidemic and for managing the epidemic during such a crisis: the degree of heterogeneity in animal movement and the dynamics of the flow of animals between farms.

The movements of cattle and swine were investigated under the conceptual framework networks and analysed using graph theory. The farms of Denmark were considered to constitute the nodes of a network and the links were the animal movements. In this framework, each farm had a sub-network of other premises to which it was linked. A premise was a farm, an abattoir or a market. If no movements of animal were registered then the network was constituted by only that farm. Otherwise, the network of the farm was linked by the movements on and off the other premises where the animals had resided. This approach allowed illustration and analysis of the four organisational levels of outputs from these registers: the animal, the movements of animals between two farms, the specific farm network and the overall industry network.

INTRODUCTION

Networks provide a conceptual framework that can express relationships between components. There are many examples of networks that are part of our everyday environment such as, the electricity distribution network and the water or sewage networks. Some networks can exist and be described at various levels and the electricity distribution network presents as a good example. Within the electricity industry, there are the main lines that distribute from the major power sources to local distribution networks, the local distribution network within the various city districts and finally the distribution of electricity within the house to the various appliances. The components of this distribution and the characteristics of the flows of electricity will differ depending on the scale at which you look at the network.

---

[1] Michel Bigras-Poulin, Faculté de Médecine Vétérinaire, Université de Montréal, St-Hyacinthe, Canada & International EpiLab, Danish Institute for Food and Veterinary Research (DFVF), Mørkhøj Bygade 19, DK-2860 Søborg, Denmark.

The network is defined largely by the components at the junction of the net and by the nature of the relationships that link these components. Networks can be studied using graph theory which provides a very rich analysis framework (Foulds, 1992)

Webb and Sauter-Louis (2002) used the network conceptual framework to investigate the contact structure of the British sheep population. They have expressed the view that contact structures have received little attention from epidemiologists even though they are very important to the understanding of disease transmission through animal contacts. In mathematical models of disease spread, animal contacts are simulated by generating stochastic movements of animals to surrounding farms within a given range. This way of modelling movements of animals and contacts assume a fair degree of homogeneity in the way that animal movements occur. It is important to verify this assumption of homogeneity. Indeed, some work has started to assess the importance of heterogeneity (Keeling et al., 2002). Very few countries have data to support the investigation of contact structure on a regular basis within the country. However, Denmark is such a country and the International EpiLab has supported a study of the animal movement data for cattle and swine.

Animal production industries possess the two basic elements necessary to define a network, nodes and links. In this epidemiological study, farming sites are the nodes or vertices, which are at the junctions in the net, and animal movements constitute the links or relations between the farming premises. In this study of animal movements, the conceptual framework used was the network, and the analytical framework was the descriptive statistics and mathematical graph theory. The conceptual frmework allows for modelling patterns of trade related to animal movements.

Trade patterns for animal movements of a specific industry are complex to study because they include many stakeholders, farming sites that are spread heterogeneously over the country, and the flow of animals between the farming sites is very dynamic. Two aspects of this complex situation are of special interest for predicting the outcome of an epidemic and for the management of an epidemic during such a crisis. They are the degree of heterogeneity in animal movement behaviour related to the specific farms, and the dynamics of the flow of animals between farms. Elucidation of these two aspects is essential to enable a fuller understanding of disease transmission by animal contacts.

There are at least two levels of organisation at which disease transmission by animal contact must be understood: within-farm level and between farm level. These levels correspond to a geographical scale of distance related to the animal contacts. In the present study, the objective was to more fully understand between farm transmission of disease through animal contact, by investigating the Danish industry's trade patterns and animal movements.

This paper is divided in two sections with the first part explaining the network conceptual framework as applied in this study. The second section shows the preliminary results of network farming premises.

NETWORKS AND GRAPHS

Networks are probably a theoretical concept that came from the fisherman's net where strings are tied together and form a lattice that is very strong and that can be very large.

These nets look like a regular series of nodes linked by strings. A graph is a theoretical entity in mathematics consisting of a set of nodes (or vertices) and a set of edges (or arcs) linking some or all of the nodes. This entity can be illustrated by a diagram, which is also called the graph. When the edges have a direction attached to them this indicates that the link is going from one node to the other, then the graph is called a directed graph and the word arc is used instead of edge.

Nodes are connected when there is a link or a series of links that can join them together. Thus, if there is a path that can be followed using edges or arcs to go from one specific node to another then the nodes are said to be connected. In a directed graph, these connections will also have direction.

The cattle or swine industry's can be considered as networks, in which commercial farms, markets and abattoirs constitute vertices and movements of one or more animals between two vertices (i.e. premises) constitute a link. The links possess a direction, since it is a movement from one premise to another. A network constitutes a graph and in this specific case a directed graph. Thus, graph theory as developed in mathematics can be used to study the network.

Trade pattern is an expression that refers to an image (or pattern) created by the trade of animals. Graphs can be used to illustrate the image of animal movements within the industry. In a directed graph, there are two major aspects of the graph that are of interest for the investigation of the graph: the shape of the diagram and the flow along the arcs.

To investigate the shape of the diagram adjacency matrices can be used (Foulds, 1992). They are n by n matrices which are a partial representation of the linkages present in a graph of n nodes or vertices. The elements of this matrix are zero when there is no path linking the nodes i and j, and are 1 when such a path exists. A path linking two nodes has a degree, which corresponds to the number of edges that have to be crossed to go from node i to node j. Thus, adjacency matrices also have a degree, which corresponds to the degree of the paths used to constitute them.

Reachability matrices are also useful. They are n by n matrices which are a partial representation of cumulative linkages present in a graph of n nodes or vertices. The elements of this matrix are zero when there is no path of degree d or less linking the nodes i and j, and are 1 when such a path exists. Thus, reachability matrices also have a degree d, which corresponds to the degree of the paths used to constitute them. They represent the union of adjacency matrices of degree d or less.

The graph of the network of the cattle industry or the one for the swine industry are huge diagrams criss-crossed by so many links that it would nearly cover the complete map of Denmark. There are subnets, which can be called farm specific networks, that represent a part of the overall network. These subnets are made up of the farming sites that send animals to other farming sites that eventually lead to the specific farming site of interest. These would be inward or outward movements. The subnets are also made up of the farming sites that received animals from this specific farming site and then send animals to other farming sites, and so on. These latter links would be the outward, or going away from, animal movements. The interest with these farm specific subnets is that they would be directly related to the risk of transmitting a pathogen through animal contact (outward) and to the risk becoming infected by a pathogen through animal contact (inward). They could be used in estimating the risk potential from a specific farming site. Friedman and Aral (2001) used a similar concept in their definition of risk-potential networks. The shape of the graphs of those networks can be illustrated by diagrams and analysed by graph theory.

The large-scale industry network is then the union of the farm specific networks of all the farming sites that constitute that industry. Thus, there are at least two levels of organisations that can be recognised, industry and farm specific.

The flow of animals within the networks can be studied for the overall network, for the farm specific networks, for the arcs and for individual animals. In animal movements, there are two distinct but related entities that must be recognised: the movement of animals and the animal that is moved. Thus, it is possible to plot a graph followed by an individually identified animal during its lifetime and it is also possible to plot a graph of the movements of animals in general. For example, a study of flow along an arc will involve the number of animals moved, the number of movements, distance and the speed of movements which is associated to the time duration between movements and during movements.

The proposed conceptual framework used has the potential to provide a four level hierarchical structure: the individual animal (for cattle only), the movement of animals between two farms (on a single day), the specific farm network, and the cattle or swine industry networks. The farm network is composed of multiple movements of animals that happen along specific arcs. Since each animal movement has a date, a farm of origin and a farm of arrival, they also have a corresponding arc which is defined by the farm of origin and the farm of arrival. A farm network is constituted of all farms and arcs that lead to this specific farm and of all farms and arcs which move away from that specific farm. In the present study, four arcs moving from and leading to the specific farm, which occurred during the observation period, were used to construct the farm network for each specific farm (paths of degree 4 or less). In the fourth level, all farms and arcs registered for the industry were considered as part of the network. The animal industry networks were not analysed *per se* in the present study, only the population of specific farm networks were analysed.

The network conceptual framework with its capacity to have shape and to support flow is conceptually rich. It can be used for in-depth investigation of the complex problem of the system of trade patterns of an animal industry. It can also be used to assess disease transmission in relation to animal contact.

MATERIALS AND METHODS

Data

Data sources were the multiple electronic registers maintained in Denmark to support agriculture. Some registers are for all agricultural activities such as the land register. In this study, only the animal industry registers were used. The first and general register was the Central Husbandry Register (CHR). This register contains general information on all farms and a farm is defined as a geographical site where animals are kept and it has a unique identification code as well as address and owner information. For each farm, there are one or more related herds. A herd is defined by animal species and usage. Each herd has a unique identifier, a code for the animal species, usage type and number of animals in the herd. This information is updated annually. If no information is received from the farm owner then the *status quo* is assumed.

Cattle

The movements of cattle were studied for the period from 1st November 2002 to 30th April 2003. For cattle, there is a specific register which contains more detailed information on the activities of the cattle industry, called the Central Cattle Register. There is also a register of animal movements. The latter register contains information on reason for movement, movement identification, cattle identification, date, and CHR identification, either for farm of origin or farm of arrival. In this register the animal is registered from the premise on the way out and, by the other premise, on the way in. Thus, there are at least two registrations for each cattle movement. The death or slaughter of each animal is also registered. For example, if the animal is slaughtered, there will be three entries in the register: the move out of the farm of origin, the move into the abattoir and finally, the move to slaughter.

From the original register data, a database was constructed which contained several data files. A data file on farm identification, farm category (commercial farm, abattoir, sale yard), number of animals by different species (dairy cattle, beef cattle, swine). A data file on individual cattle movements, which contained animal identification, reason for movement, premise of origin identification, date of departure, premise of arrival identification, and date of arrival. A data file for daily movement of animals between two premises contained: identification of premise of origin, identification of premise of arrival, date, number of animals involved, location of premise of origin, location of premise of arrival, farm type of premise of origin, farm type of premise of arrival, and the Euclidian distance between the two geographical locations. A data file for specific farm network information which contained data on premise identification, farm type, farm location, number of dairy cattle, number of beef cattle, number of swine, number of premises adjacent from that specific premise in one arc, number of premises adjacent from that specific premise in 2 arcs, number of premises adjacent from that specific premise in 3 arcs, number of premises adjacent from that specific premise in 4 arcs, number of premises that could be reached from that specific premise in 1 to 4 arcs, number of movements of cattle on the specific farm network, sum of distances covered by the network, average distance of an animal movement within the network, and number of cattle moved in the network.

Swine

The movements of swine were studied for the period from 30th September 2002 to 22nd May 2003. For swine, a register linked to the CHR register is maintained with a large quantity of specific data on swine movements in Denmark. In the present study, the register of domestic swine movements was used. This register contained the following data: date of movement, hour of movement, identification of premise of origin, identification of premise of arrival, number of pigs in lot and truck identification. If a truck picks up many lots of animals on one route, this information was not used in this study. Thus, contacts of swine from different lots in a truck were not analysed.

From the original register data, a database was constructed using several data files. It contained a data file of farm identification, farm category (commercial farm, abattoir, sale yard) and number of animals of different species (dairy cattle, beef cattle, swine). A data file for daily movement of animals between two premises contained information on identification of premise of origin, identification of premise of arrival, date, number of animals involved, location of premise of origin, location of premise of arrival, farm type of premise of origin, farm type of premise of arrival and Euclidian distance between the two geographic locations. A data file for specific farm network information consisted of data on premise identification, farm type, premise location, number of dairy cattle, number of beef cattle, number of swine, number of premises adjacent to that specific premise in one arc, number of premises adjacent to that specific premise in 2 arcs, number of premises adjacent to that specific premise in 3 arcs, number of premises adjacent to that specific premise in 4 arcs, number of premises that could reach that specific premise in 1 to 4 arcs, number of movements of pigs on the specific farm network, sum of distances covered by the network, average distance of an animal movement within the network, and number moved in the network.

Data analysis

The analysis of the data was completed separately for cattle and swine. They were each analysed at the specific level of organisation relevant to these animal movements.

The movement of individual cattle was described using standard descriptive statistics. The movements of cattle and swine were also described in a similar fashion. The description was completed according to farm type.

Farm specific networks were illustrated using the software, PAJEK (Anon., 2003). The networks were built using computer programs developed specifically to achieve this task. The programs were developed using the M computer language from Caché Intersystems version 4.0. The programs produced a three degree directed graph from a specific farm or going to the specific farm. The resulting network was written in a format that can be read by the PAJEK software for illustration purposes (Anon., 2003). This software allows the user to view the resulting specific farm network.

Network analysis was completed at the industry level according to graph theory methods using adjacency matrices of degree one to four and reachability matrix including four degrees. Descriptive statistics were then used to present the results. Specific programs were developed using the M language to calculate the adjacency and the reachability matrices.

Geographic distances were calculated using the Euclidian distance from the UTM zone 32, EUREF 89 (WGS84), coordinates.

RESULTS

Based on preliminary results, individual cattle rarely moved in the six-month observation period. The movements of cattle between two specific commercial farms did not occur very often during the six-month observation period. This indicates an extremely skewed distribution with most premises having a small network of movements and a small proportion of premises with large networks.

DISCUSSION

Data validation is often a problem because the analysis and description of trade patterns required the construction of many data files that originate from various data sources and it was difficult to follow the trail of individual animals or movements through the data files. This construction produced missing values along the way and thus for some variables there can be a large number of missing values. This is especially true when flows are to be studied. For that reason, flows of animals within networks were not investigated but such an investigation would be of great scientific and practical interest to assist in further understanding disease transmission.

In the available data, it was not possible to take into account the movements of trucks that carry out these animal transfers as truck routes were not available. This could be important because the truck itself becomes an epidemiological unit, within which disease transmission can occur. Animals can be taken by the same truck during the travel route from different locations and then unloaded at different locations, but some animals will share the same crowded space for some time, thereby facilitating disease transmission. Furthermore, the time from date of departure to date of arrival can be up to three days in the cattle movement registry, making it impossible to record errors or to check if the animals had actually spend all that time in transit.

The animal movements can be studied by using the movement itself, as an epidemiological unit instead of an animal being moved. These two different epidemiological units (animal vs. movement) illustrate a different story. The movements of one specific animal can tell of the impact of movement on the career of an individual animal. The movement used as the epidemiological unit tells the story of the movements themselves. The latter is indicative of the flow of animals within the cattle industry.

The graphs of the specific farm network epidemiological units are directed graphs since the movement of animals always has a direction. The farms or premises are the vertices of the graphs and the movement of animals define the arcs. There can be many movements of animals that would define the same arc. Thus, on one arc there can be many transports of animals and this is indicative of variation in the flow of movements of animals. This flow would be defined by the number of movements, the number of animals for each movement and the chronology of the movements. Analysis of the flow can indicate speed or rate of movements and changes of speed. Rates would be the number of movements on the arc by time unit or for the number of animals moved on the arc by time unit. In graphs, there are also paths which are indicative of the relationships among arcs or the way in which a pathogen can travel from one farm to another, even if it takes more than one movement of animals. In a path, a link between two vertices or farms in one movement of animals is called a step. The adjacency matrices indicate the number of vertices that could be joined to a specific vertex using a specific number of steps. The number of steps is then called the degree of the adjacency matrix. The reachability matrices indicate the number of vertices that could be joined to a specific vertex using up to a specified number of steps. The adjacency and reachability matrices in a directed graph are dual since there are two possible directions where the inward movements going to the specific farm and the outward movements going away from the specific farm.

Networks

The existence of an individual animal identification for cattle made it possible to look at animal movements on an individual animal basis. Visual assessment of the example farm shown in Fig. 1 illustrates the network of premises reached through the trade patterns of that farm. The power of the picture when compared to Fig. 2 shows the impact that the diagrams of these graphs can provide to the user.

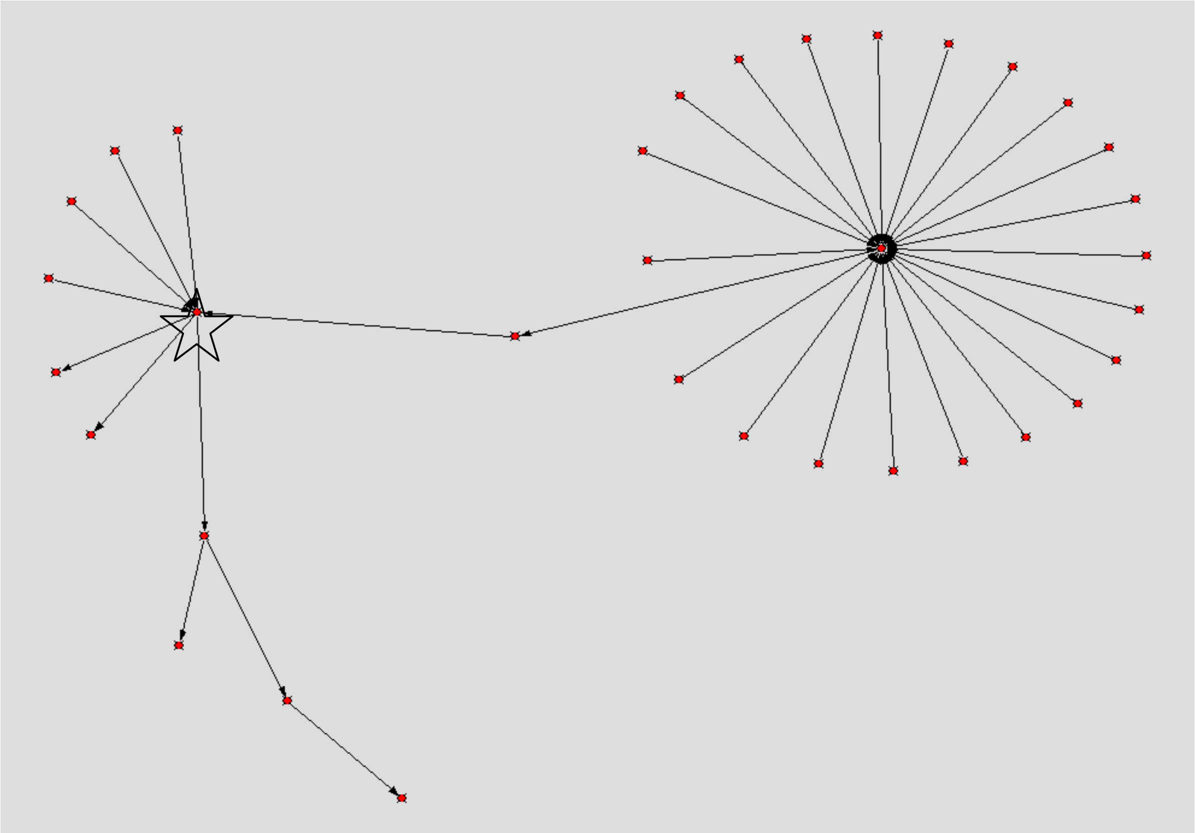


Fig. 1 A commercial beef farm (shown by the star) with 90 cattle, and the movements of cattle that reached other premises in up to 3 links inward and outward of the specific farm.

An important heterogeneity in trade patterns observed between the cattle farms, as well as for markets and abattoirs, seems to be present in the preliminary results. The risk potential of disease transmission through animal contact is much greater for these premises. This risk potential will vary from disease to disease and even between strains of pathogen. The above discussion indicates that an assumption of homogeneous animal movements throughout the industry is not acceptable. The heterogeneity between the various farms must be taken into account.

The heterogeneity in trade patterns was also observed between the pig farms, as well as for markets and abattoirs. The risk potential of disease transmission through animal contact is much greater for these latter premises.

The classification of pig or cattle farms according to their specialised activities would make the understanding of trade patterns much clearer. Also, the risk potential will vary from disease to disease and even from one strain of pathogen to another. Thus, the characteristics of the networks should be weighted differently for the different situations. The above discussion and preliminary results indicate that an assumption of homogeneous animal movements throughout the industry is not acceptable. The heterogeneity between the various farms must be taken into account.

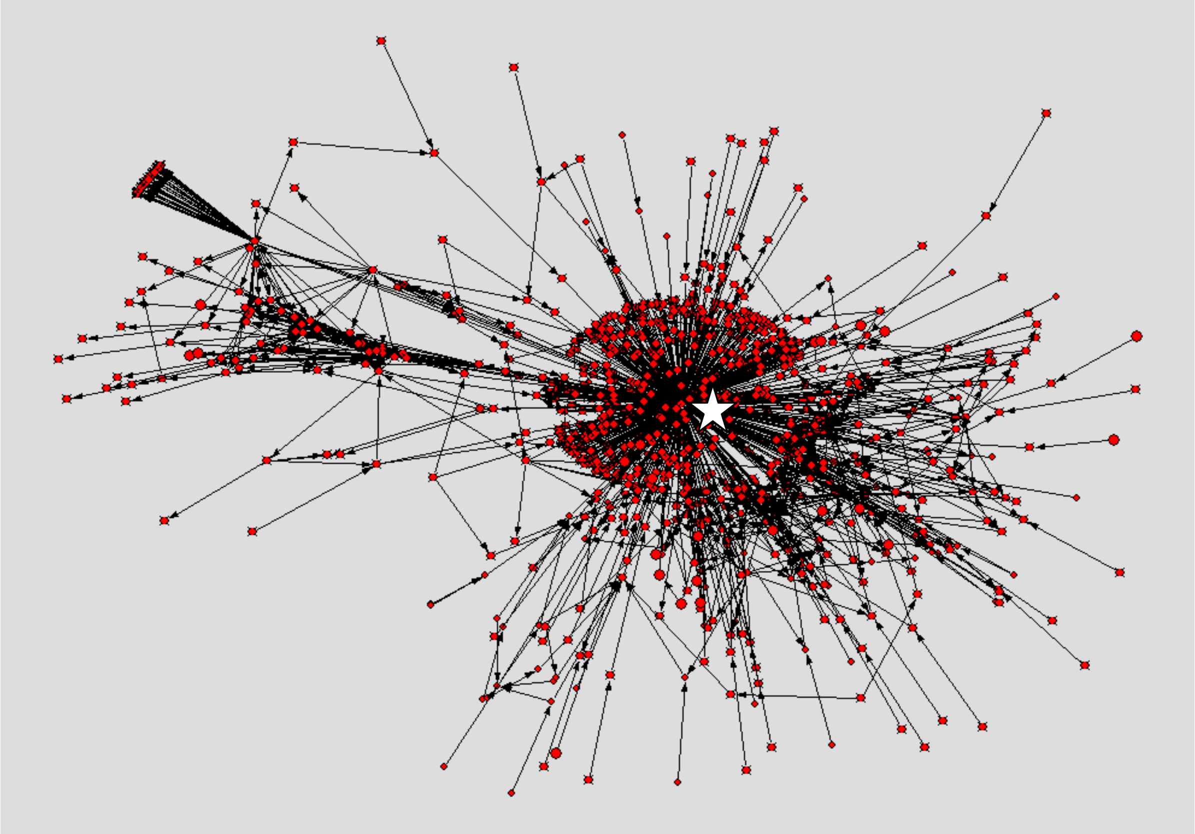


Fig. 2 A market (shown by the star) with animal movements that reached other premises in up to 3 links inward and outward of the specific market.

Impact of animal movements on a potential epidemic of foot-and-mouth disease (FMD)

The key issue of this part of the study was to evaluate the degree of between farm heterogeneity in the movement of animals. Graph theory was used to compare the networks related to animal movements between cattle and swine farms. The results indicated an important degree of species specificity and an asymmetric and important variation between farms in relation to animal movements. The farm specific networks showed a tendency to be very large for 5-10% of premises. This indicates that heterogeneity has to be taken into account when managing an epidemic crisis.

Certain strains of the FMD virus have the capacity to spread very efficiently by animal contact. The virus also has a 4-9 day period, in which the animal is contagious but where no clinical signs can be observed. This period can even be longer for certain virus strains because the clinical signs can be very hard to detect. The period between the occurrence of the index case and detection of the epidemic can be relatively long (approximately 3 weeks in the case of the UK 2001 FMD epidemic) thereby allowing for further spread by animal movement. Because of this detection problem at the onset of the epidemic, it is very important to use adequate levels of appropriate human resources to achieve rapid control of the epidemic. To accomplish effective control, it is essential to know the risk potential of the index farm or farms and to have the ability to trace animals at as local a level as necessary.

The network analysis used, especially if the knowledge is made available through a geographic information system, can provide a means to achieve the trace history and risk potential level of the premises involved with the index case(s). The graphs produced are farm specific and are at a detailed enough scale to show geographical location and farm information.

Mathematical models of disease spread generate animal movements based on farm size using a constant stochastic animal movement algorithm and a fixed area within which these movements are possible. These are constraints, which assume a large degree of homogeneity in animal movements for any farm. This will average out over the course of the epidemic, if many iterations are completed. For this reason, the simulation model is useful for the evaluation of various strategies, given that the characteristics of the viral strain are known or presumed known. It is a very efficient preparedness tool. On the other hand, this assumption of homogeneity makes the simulation model much less useful in helping to control an epidemic. The network approach proposed in this project is likely to be more appropriate to do this because it can incorporate the heterogeneity associated with animal movement.

The network approach is dependent on regularly updated animal movement and on a database with valid and complete data. The update time is a crucial characteristic of a crisis management system based on the network approach. It would be very useful where animal movements from the index case(s) were readily available. For prolonged management usage throughout the crisis, the movement of animals must be updated in real time. The route of trucks used in animal transportation could also be registered. This should be considered since it is likely that an animal standstill will be harder to enforce in the future (Webb & Sauter-Louis, 2002).

ACKNOWLEDGEMENTS

The research was funded as a project of the International EpiLab (93S-2465-Å01-01269) and was completed at the International EpiLab, Denmark. The funding came from various sources within the government and animal industries of Denmark. The authors wish to thank Mette Marie Larsen for her help with the data, Anette Boklund from the Danish Bacon and Meat Council and Flemming Skjoth from the Danish Cattle Associations.

REFERENCES

Anon. (2003). PAJEK, program for large network analysis. University of Ljubljana, Slovenia. HTTP://vlado.fmf.uni-lj.si/pub/networks/pajek

Foulds, L.R. (1992). Graph theory applications. Springer-Verlag New York Inc.

Friedman, S.R. and Aral, S. (2001). Social networks, risk-potential networks, health and disease. J. Urban Health 78, 411-418

Keeling, M.J., Woolhouse, M.E.J., Shaw, D.D., Matthews, L., Chase-Topping, M., Haydon, D.T., Cornell, S.J., Kappey, J, Wilesmith, J. and Grenfell, B.T. (2002). Dynamics of the 2001 UK foot and mouth epidemic: Stochastic dispersal in a heterogeneous landscape. Science 294, 813–817

Webb, C.R. and Sauter-Louis, C. (2002). Investigations into the contact structure of the British sheep population. Proceedings of the Society for Veterinary Epidemiology and Preventive Medicine, Cambridge, 10-20.

THE EFFECT OF REGULATIONS ON THE CONTACT STRUCTURE OF THE DUTCH CATTLE SECTOR

A.G.J. VELTHUIS[1]

SUMMARY

After the FMD outbreak in the Netherlands in 2001, the Dutch government implemented some regulations with the goal of reducing the number of animal contacts between farms and consequently the risk of introduction and the spread of highly contagious animal diseases in the future. In this study, the effectiveness of these regulations was investigated. Identification and Registration data of the Dutch cattle sector were used to compare 1) the number of animal and transport movements before and after the FMD outbreak, and 2) the contact structure between different farms and holdings of the Dutch cattle sector before and after the FMD outbreak. It could be concluded that the number of animal movements decreased, the number of transport movements increased and that the contact structure changed after the FMD outbreak. A lot of these changes are due to the new regulations but they are also due to the decrease in the number of active farms after the FMD outbreak. Whether the spread of FMD or other infectious diseases will be reduced with this new contact structure could not be concluded by this study.

INTRODUCTION

On 21st March 2001, foot and mouth disease (FMD) was diagnosed in four cows on a farm in Olst, the Netherlands. This highly contagious disease occurred in the Netherlands, despite all precautions, after the United Kingdom. In total, 26 farms were infected and 2763 farms were culled preventively during the outbreak (Bouma et al., 2003). The Department of Agriculture, Nature Management and Food Quality set the FMD crisis plan in motion immediately after the first farm was diagnosed positive. A temporary movement ban was laid down for the whole of the Netherlands which applied to: all livestock and poultry, vehicles for the transport of livestock and poultry, semen, ova and embryos from biungulates, milk and feed materials. Farm premises were off-limits to visitors except in prescribed emergency situations. The plan also prescribed that all biungulates (most commonly sheep, goats, pigs and cattle) had to be killed and destroyed as soon as possible after FMD has been confirmed on a farm. Susceptible animals on farms situated within one kilometre of an infected farm were pre-emptive culled and a three kilometre protection and ten kilometre surveillance zone were implemented. The FMD crisis plan laid down other temporary regulations, such as import and export restrictions.

---

[1] Annet Velthuis, Business Economics, Wageningen University, Hollandseweg 1, 6706 KN Wageningen, The Netherlands. Email: Annet.velthuis@wur.nl.

All regulations were aimed at reducing the spread of the FMD virus. Most of them were withdrawn at the end of the outbreak but some became were retained albeit slightly reformulated. The aim of some of the irreversible regulations was to change the contact structure of the Dutch cattle sector in such a way that the number of animal contacts between the different farms and holdings in the cattle sector was minimised. This was in order to reduce the spread of highly contagious animal diseases in future outbreaks.

The first regulation, the so-called 'live gathering' regulation, prohibits the gathering of biungulates for a period shorter than 30 days. Exceptions are gathering at certified collecting centres and slaughterhouses or gathering on one truck, if all animals are transported to one farm, livestock collecting centre or slaughterhouse. The second regulation the so-called 'thirty days' regulation prohibits the movement of biungulates off a farm within 30 days after a biungulate has been moved on to that farm. Exceptions are the sale of pigs, sheep that graze temporarily on other farms, and biungulates that are transported to a slaughterhouse via a collection centre. These two regulations caused many problems for the livestock sector and there was a lot of protest. Therefore, the government and the livestock sector agreed that the regulations would be evaluated one year after implementation.

This study was based on the evaluation (Greutink et al., 2002). Three questions were formulated for this evaluation. First, is it possible to fulfil these regulations and are they fulfilled properly in practice? Second, have these regulations changed the contact structure of the livestock sector? Third, will these regulations reduce the spread of highly contagious animal diseases in future outbreaks? The second question has been addressed in this study. For this, only the cattle sector has been under investigation, because good data of individual cattle movements were available. The research question was therefore: did the regulations result in less animal contacts between the different farms and other holdings in the Dutch cattle sector? With the help of Identification and Registration (I&R) data of the Dutch cattle sector we compared 1) the number of animal movements, 2) the number of transport movements and 3) the contact structure between different farms and other parties of the Dutch cattle sector before and after the FMD outbreak.

MATERIALS AND METHODS

Description of I&R data

Data from the National Identification and Registration system were used. In table 1 an overview of the notifications used is given. Each notification consists of the notification code, the ID number of the animal, the date, and the unique farm identity number (UBN) of the farm or holding to which the notification applies. All notifications dealing with animal movements (all except the birth notifications with code 10) were used for the analysis.

Definition of periods

To compare the effect of the applied regulations, data from before and after the FMD outbreak were analysed and compared. The pre-outbreak period, which is called '2000' in this paper, was a period of 9 months and ran from the first of May 2000 until the first of February 2001. The post-outbreak period, which is called '2002' in this paper, was a period of 12 months and ran from the first of September 2001 until the first of September 2002. The pre-outbreak period did not comprise a whole year because data before the first of April 2000 were partly deleted from the system. For the comparison of the total number of movements between the two periods, the number of movements in period 2000 was scaled to 12 months by multiplying all numbers with 12/9.

Table 1. Codes and descriptions of the notifications used in the Dutch I&R system for cattle and the number of notifications[1] used and the percentage of the total in the defined pre- and post- FMD periods.

			2000[3]		2002[4]	
CODE	DESCRIPTION	TYPE[2]	NUMBER	%	NUMBER	%
10	Birth		749,386	8.7	772,644	7.4
18	Re-import	→●	350	0.0	285	0.0
19	Birth and send off	●→	494,205	5.7	967,184	9.2
20	Send off	●→	2,342,490	27.2	3,079,601	29.4
21	Export	●→	74,335	0.9	173,396	1.7
29	Send off of IKB[5]-certified animal	●→	161,741	1.9	326,217	3.1
30	Arrival	→●	1,587,535	18.4	1,607,342	15.3
31	Import	→●	508,225	5.9	468,528	4.5
32	Transit / trade	●→&→●	846,609	9.8	701,576	6.7
40	Death	●→	106,992	1.2	249,931	2.4
41	Slaughter	→●	1,540,872	17.9	1,951,471	18.6
	Total		8,607,484	100.0	10,480,944	100.0

Including double notifications

For one movement a ‘sending off (dot then arrow)’ and a ‘receiving (arrow then dot)’ notification is needed.

Covers a period of more than 9 months from 1st April 2000 until 1st February 2001.

Covers a period of more than 12 months from 1st August 2001 until 1st September 2002.

IKB (Integrale Kwaliteits Bewaking) is a certification scheme for quality meat products.

From notification to animal movement

Each notification dealt with one event: sending off or receiving the animal. Thus, for an animal movement – within the Netherlands – two notifications of two different farms were needed. To complete an animal movement the two successive notifications were found and coupled. This coupling was done in different steps. First, all successive notifications that consisted of the same animal ID and date were examined closely. Second, when available, the transit notifications (code 32) were coupled to each other. This meant that an animal had been on two cattle-collection centres or trade farms in a row. Third, the transit notifications were coupled to other sending-off or receiving notifications. Fourth, all other notifications were coupled. Used notifications were blocked directly after the coupling, except for the transit notifications, which were blocked after the second time they are used in a movement. These steps were repeated for each difference in arrival and departure date up to a 14 day difference. Coupling of the import and export notifications was not necessary, because foreign farms were not included in the Dutch I&R system. These notifications were directly transformed into movements to and from abroad.

Five movement types can be differentiated: for live gathering, for slaughter, for destruction, import or export. Movements in which code 41 is used are ‘for slaughter’, in which code 40 is used are ‘for destruction’, in which codes 18 and 31 are used are import movements and in which code 21 is used are export movements. All other movements are ‘for live gathering’.

From animal movement to transport movement

A transport movement included all animal movements that were registered at the same departure and arrival dates from and to farms or holdings with the same UBN. More transport movements were assumed if more than 40 adult cattle, 100 veal calves ready for slaughter or 300 very young veal calves were transported on the same date from and to the same holdings.

Calculation of the distance

For all movements within the Netherlands the direct distance between the two farms or holdings was estimated. For each UBN the x and y co-ordinates were identified. If (x1,y1) are the co-ordinates of the ‘sending off’ farm and (x2,y2) the co-ordinates of the ‘receiving’ farm, then the direct distance between the two was calculated as follows:

$$\text{distance} = \sqrt{(x2 - x1)^2 + (y2 - y1)^2}\ .$$

This calculated distance represented the length of a straight line between the two farms and was therefore the minimum movement distance. Thus, the actual distance of the route via the road was bigger than the calculated.

Farm types

Each farm or holding that trades, keeps or processes cattle in the Netherlands should have a unique farm identity number (UBN). In the two periods, 67305 different UBNs or farms have notified the Dutch I&R-system. Although it is possible that more than one UBN is located at one address (12.3%) it is assumed that each UBN represents one single farm or holding.

Registration of the type of farm or holding for each UBN in the Dutch I&R-system was available but the data were not up-to-date and were incomplete. Therefore, the farm type had to be identified. This was done with help of another database of the Dutch I&R-system. This database contained the number of animals in three different age classes on four predefined dates from the period of May 1st 2000 until February 1st 2000, for each UBN. Based on these numbers and on the number and type of notifications made in the 2000 database, a farm type was defined for each UBN. In Table 2 the different farm types and the descriptive statistics based on the period 2000 are given.

Table 2. The farm types with description, the number of farms per type for both periods, the average number of animals present in three age classes and the average number of animals that were send off from that farm or have arrived on that farm type, based on the period 2000.

		# UBNS		# ANIMALS PRESENT ON FARM						AVERAGE # ANIMALS MOVED			
TYPE	DESCRIPTION	2000	2002	<1 Y	STD.	1-2 Y	STD.	>2 Y	STD.	FROM	STD.	TO	STD.
Da	Dairy	27661	27370	20.6	13.4	20.0	12.6	60.7	33.1	32.6	18.3	2.5	6.6
Da/Be	Dairy and beef[1]	327	319	29.0	22.0	30.2	21.0	29.9	36.0	26.8	22.9	12.3	17.9
Da/Tr	Dairy and trade	171	166	6.4	9.1	9.7	12.0	58.3	49.5	60.2	40.1	20.9	20.0
Yo	Young stock raising[2]	579	524	13.9	23.0	23.1	29.4	8.1	13.3	10.7	10.3	7.7	11.1
Be	Beef[1]	3768	3380	191.4	243.2	10.4	24.1	4.7	12.1	17.8	20.2	58.6	104.9
Tr	Trade[3]	3087	2642	9.2	40.9	10.1	21.5	13.3	21.1	24.4	48.8	29.3	402.9
Be/Tr	Beef and trade	128	119	309.7	326.7	7.7	39.3	11.2	37.0	31.2	37.3	87.0	101.0
Su	Suckling cows[4]	2361	2316	8.1	7.3	8.0	7.4	20.1	16.8	12.5	16.1	5.3	13.6
Cc	Cattle collection centre	20	6	0.0	0.0	0.1	0.5	0.1	0.5	1839	3448	2709	5398
C-Cc	Certified cattle collection centre[5]	77	61	23.4	89.6	7.6	20.4	10.7	21.1	370.4	1160	2127	5473
Others	Not[6]	20828	16089	1.8	2.4	2.1	3.0	3.5	4.4	2.9	3.5	1.3	2.4
Sl	Slaughterhouse[7]	412	296	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	586.3	2792
New	Farms that did not own or trade animals in period 2000	0	1808	7.3	67.0	1.0	0.0	1.4	8.1	5.1	12.9	11.7	68.4

1) veal calves or bulls
2) sometimes combined with beef
3) import en export included
4) sometimes combined with beef and trade
5) certification was obliged from 1 January 2002
6) E.g. small farms that own <10 cows, cattle markets and cattle shows
7) including ‘at home’ slaughters

It was assumed that the farm type did not change between the two periods. Only if a farm stopped its activities was it excluded from the calculations. If a non-active farm of 2000 became active again in 2002, it was classified as a 'New' farm in 2002 (see Table 1).

RESULTS

From notification to animal movement

A total of 5.8% of the pre-outbreak notifications (2000) and 12.7% of the post-outbreak notifications (2002) could not be coupled to an animal movement. One reason that the percentage in the post-outbreak period was higher than the post-outbreak period was that it was no longer possible to use the combined 'birth and send off' notification (code 19) as a 'send off' notification as from 2001. The affected animals were therefore also treated as a separate 'send off' notifications.

Animal movements per month

The start and the end date of a movement could differ and fall in two different months. Therefore, the allocation of movements to different months was based on the start date of the movement, which was the date of the 'send off' notification. The import movements were based on the end date, because the start date was unknown. The number of animal movements per month and the average difference between the two periods are given in figure 1 and table 3, respectively.

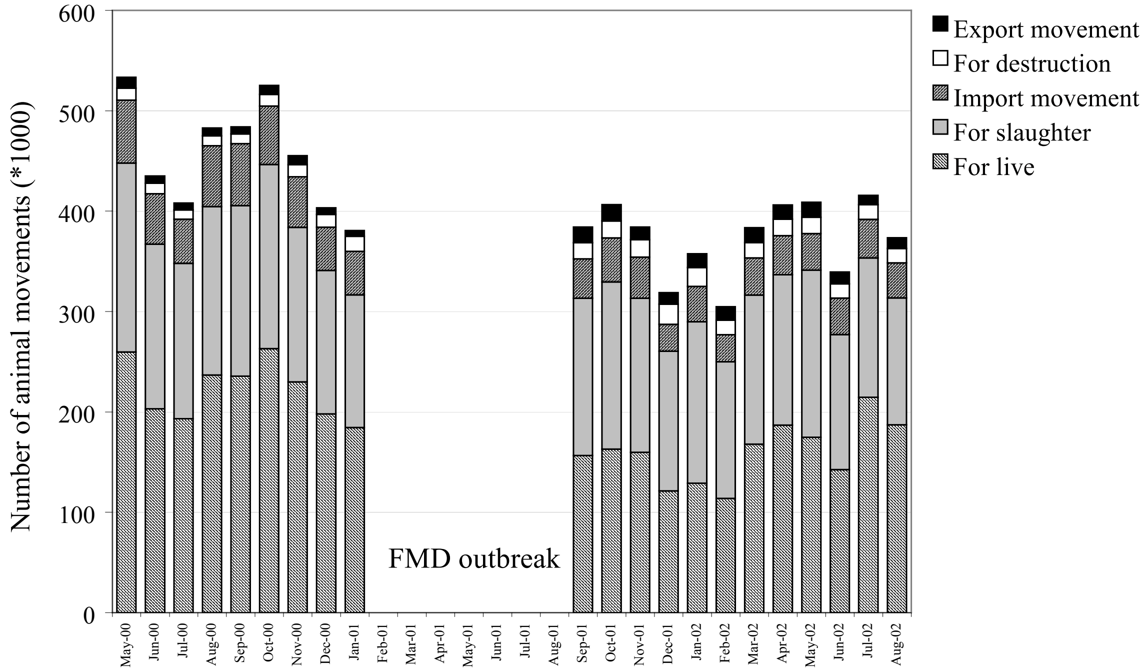


Fig. 1 Number of animal movements per months from May 2000 to August 2002.

The number of animal movements decreased significantly after the FMD outbreak with 82852 movements per month. This difference was for the greater part attributed to movements of live animals for collection (62879 movements). This decrease might have been caused by the new regulations in combination with the decrease in the number of farms (see table 2).

The number of animals moved for this purpose and the number of imported animals per month decreased significantly after the FMD outbreak, with 62879 and 16441 movements per month, respectively. The number of exported animals per month and the number of animals going for destruction each month increased significantly with 5480 and 4833, respectively. The latter was probably due to an administrative "catch-up" with notification of animals that had been culled during the FMD outbreak. Also slaughterhouses started to notify that cows which were not suitable for human consumption were sent from the slaughterhouse to the destruction plant in 2002.

Table 3. The average number of animal movements per month, the standard deviation and the difference in number of movements between the pre-outbreak period (2000) and the post-outbreak period (2002).

	2000		2002		DIFFERENCE
MOVEMENT TYPE	# / MONTH	STD.	# / MONTH	STD.	# / MONTH
For live	222591	27008	159712	29587	62879*
For slaughter	162015	15205	148169	13171	13846 .
Import	52642	8009	36201	5063	16441*
For destruction	11440	1314	16273	1838	-4833*
Export	7773	1471	13253	2124	-5480*
Total	456460	49334	373608	36509	82852*

* Significant with α=0,05 based on a student t-test.

<u>Contact structure based on animal movements</u>

The contact structure based on animal movements between the different holdings in the Dutch cattle sector is summarised in the contact matrix of appendix A1. The difference in the number of movements is calculated by scaling the number of movements in 2000 to one whole year (x 12/9) and distract it from the number of movements in the period of 2002. In appendix A2 the difference between the two periods is given. In the following paragraph only big differences are discussed.

The total number of animals moved decreased after the FMD outbreak with 1,246,302. Dairy farms (Da) sending off more and receiving fewer animals in 2002. Many more animals were moved from dairy farms directly to beef farms, traders and slaughterhouses and far less from dairy farms to the collection centres. The number of dairy farms decreased after the FMD outbreak with 291 farms.

The total number of animals transported to beef farms (Be) and sent off from beef farms decreased a lot after the FMD outbreak. A lot of animals transported to a beef farm come directly from dairy farms or foreign countries, whereas in 2000 most animals are coming from collection centres. The number of beef farms has decreased also with 388.

Traders moved fewer animals to and from their farms in period 2002 compared to 2000. Traders imported a lot fewer animals and sold many less animals to beef farms. The total number of trade farms decreased by 445.

The activity of collection centres (certified or not) decreased a lot after the FMD outbreak. Notably, the flows from dairy farms to collection centres and from collection centres to beef farms are much smaller.

Furthermore, the total number of imports decreased a lot, which also applies to the number of animal slaughtered. A total of 1808 holdings that were not active in the 2000 period started up again after the FMD outbreak and moved a lot of animals.

Transport movements per month

The number of animal movements per month and the average difference between the two periods are given in figure 2 and table 4, respectively.

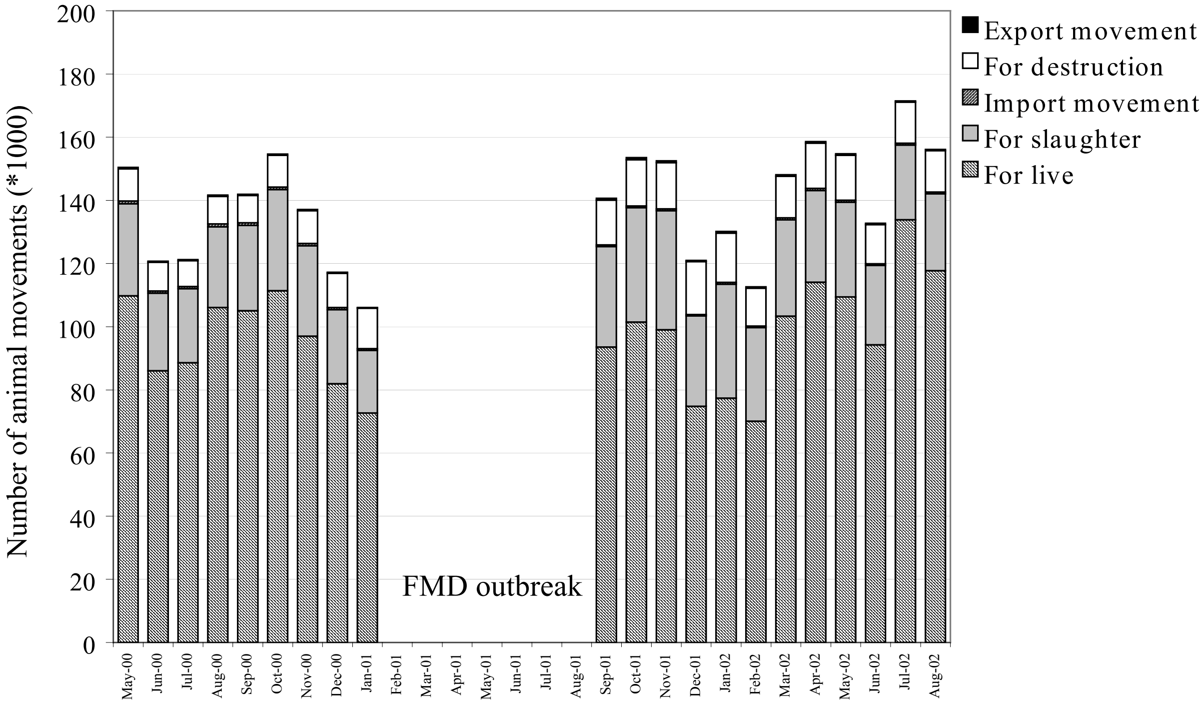


Fig. 2 The number of transport movements per months from May 2000 to August 2002.

The number of transports increased from 132329 to 144343 per month. The number of live transports was not significantly different between both periods. However, the number of transports to the slaughterhouse, and the destruction plant increased significantly. The number of imports decreased significantly, whereas the number of monthly exports increased significantly.

Contact structure based on transport movements

The contact structure based on transport movements between the different holdings in the Dutch cattle sector is summarised for both periods in the contact matrix of appendix B1. The difference in the number of transport movements has been calculated in the same manner to the difference in the number of animal movements and is given in appendix B2. In the following paragraph only the big differences are discussed.

Table 4. The average number of transport movements per month, the standard deviation and the difference in number of movements between the pre-outbreak period (2000) and the post-outbreak period (2002).

	2000		2002		DIFFERENCE
MOVEMENT TYPE	# / MONTH	STD.	# / MONTH	STD.	# / MONTH
For live	95385	11485	99088	18754	3703 .
For slaughter	25972	3036	30234	4651	42628*
Import	723	109	539	72	-184*
For destruction	9907	970	14027	1364	4120*
Export	342	62	455	87	113*
Total	132329	14288	144343	17195	12014 .

* Significant with α=0,05 based on a student t-test.

The total number of transports increased in 2002. The number of transports coming from dairy farms caused a big part of this increase. The number of transports from dairy farms to beef farms, trade farms, slaughterhouses and the destruction plant increased a lot whereas the number of transports from dairy farms to cattle-collection centres decreased in 2002. The number of transports to the dairy farms decreased in 2002.

The number of transports from the beef farms to the slaughterhouse decreased. The number of transports to the beef farms increased in 2002. Most of them were coming directly from the dairy farms, whereas the number of transports originating from collection centres decreased a lot in 2002. The number of transports between traders mutually and between traders and collection centres decreased a lot in 2002. Also the number of transports from dairy farms to traders decreased. A total of 1808 farms classified as empty became active in 2002. The actual holding types were not revealed, but a lot of transports to these farms were coming from dairy farms and most transports coming from these farms went to all different kinds of farm.

Number of contacts per farm

The average number of different farms or holdings that had contact with one typical farm of a specified type has been calculated for both periods. In table 5 the average number of contact farms, subdivided for farms that delivered to or received cattle from a typical farm for both periods, is given.

The average number of different receiving farms, to which a typical dairy farm is moving cattle, increased from 14.08 in 2000 to 26.13 in 2002. The number of different delivering farms (that deliver cattle to a typical dairy farm) remained the same for both periods. The number of delivering farms for a typical beef farm increased a lot, from 56.77 in 2000 to 169.56 in 2002. This was due to the fact that most calves were delivered by dairy farms directly and not via collection centres, which were less active in 2002. The average number of delivering farms to a cattle collection centre decreased a lot from 294.29 in 2000 to 46.84 in 2002.

Table 5. The average number of farms (and standard deviation) that had contact with one typical farm of that farm type, subdivided to delivering and receiving farms for both periods, 2000 and 2002, respectively.

	DELIVERING FARMS				RECEIVING FARMS			
	2000		2002		2000		2002	
TYPE	#	STD.	#	STD.	#	STD.	#	STD.
Da	3.07	4.64	3.37	7.97	14.1	8.35	26.1	13.1
Yo	5.33	7. 03	4.75	6.43	5.75	5.15	5.94	4.97
Da/Be	9.00	9.87	8.94	14.7	11.9	8.46	13.3	11.4
Be	56.8	93.7	169	247	4.30	4.08	4.38	3.41
Da/Tr	12.1	11.1	15.8	31.8	25.1	16.7	31.0	22.2
Tr	14.7	101	19.6	152	10.7	20.0	9.27	12.8
Be/Tr	73.2	91.2	222	223	5.71	6.61	5.50	6.06
Su	5.55	8.55	5.89	11.9	6.43	5.31	8.54	7.10
Cc	2150	3355	202	306	625	1063	24.0	24.0
C-Cc	686	1197	923	1089	178	399	48.4	68.6
Cc & C-Cc	1087	2076	857	1062	294	662	46.8	66.8
Others	2.19	2.17	2.87	10.9	2.77	2.22	3.27	3.17
Sl	237	1048	415	1638	0.00	0.00	1.00	0.00
New	1.14	0.53	13.7	68.0	1.30	0.92	3.79	6.65

Distance of transports

The cumulative distribution of the direct distances that were covered by the transports for "live gathering" is illustrated in figure 3. From this figure it can be seen that the distance of the transports in the period 2002 was bigger than in period 2000.

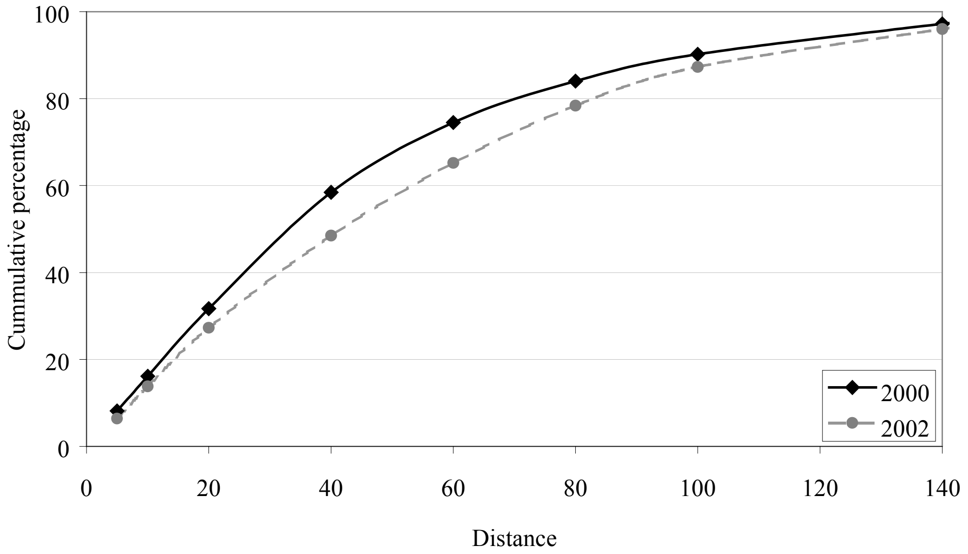


Fig. 3 Cumulative distribution of the transport distances for live for the two periods 2000 and 2002.

DISCUSSION

The research question was; “Did new regulations on animal movements change the contact structure of the Dutch cattle sector?” The relevant regulations were aimed at reducing the spread of possible highly contagious animal diseases by reducing the number of animal contacts between farms or other cattle holdings. The regulations were briefly a regulation that prohibited the collection of live cattle on cattle markets or cattle collection centres, and a regulation that prohibited a farmer to move cattle live from his farm within 30 days after cattle had moved on to his farm.

Data of the Dutch Identification and Registration system were used to answer this question. The answer to the research question is that the number of animal movements decreased, the number of transport movements increased and that the contact structure has changed after the FMD outbreak.

The most important changes were that the number of transports from dairy farms to cattle collection centres decreased significantly, and that the number of transports from dairy farms to beef farms (including veal calf farms) increased significantly. The total number of transports towards and from cattle collection centres decreased significantly after the FMD outbreak. Each dairy farm delivered cattle to more different farms after the FMD outbreak; the number of receiving farms per dairy farm was 14.1 before, and 26.1 after the FMD outbreak. Furthermore, more different farms delivered cattle to each beef farm; the number of delivery contact farms was 56.8 before and 169.6 after the FMD outbreak. The number of delivery and receiving contact farms to and from one average cattle collection centre decreased from 1087.0 to 856.9 and from 294.3 to 46.8, respectively.

The trading of cattle has become much less attractive due to the new regulations. This is illustrated by the fact that the number of trading farms decreased a lot and that the number of animal movements and transport movements via trade farms decreased. The activity of cattle collection centres also decreased a lot. Only gathering of cattle was allowed on certified cattle collection centres in 2002.

Summarising, it can be concluded that the number of animal movements decreased and that the contact structure changed after the FMD outbreak. A lot of these changes were due to the new regulations. However, some effect can also be attributed to the decrease in the number of active farms after the FMD outbreak. Whether the spread of FMD or other infectious diseases will be reduced with this new contact structure can not be concluded at this point.

Another reason for publishing the contact structure of the Dutch cattle population is to provide data for epidemiological studies on the spread of animal diseases. Many of the existing models developed to explore disease transmission and control options assume a homogeneous mixing of farms or take on an explicit spatial structure but assume a random contact structure. A good understanding of the cattle contact structure would enable models to capture the true heterogeneity in the system and allow more realistic comparison of control strategies than is currently possible (Nielen et al., 1996a; Nielen et al., 1996b). In another study carried out for this evaluation the interFMD model has been used, based on the contact structures of 2000 and 2002, to simulate FMD outbreaks in the Netherlands. The results of this study have only been published in a report (Greutink et al., 2002).

The need for a good understanding of the contact structure of farm animal population became imperative during the FMD outbreak in Great Britain, where efforts to use mathematical models to forecast the spread of the FMD virus were hampered by a lack of information on this contact structure (Webb et al., 2002)

ACKNOWLEDGEMENTS

This study was funded by the Dutch Ministry of Agriculture, Nature Management and Food Quality.

REFERENCES

Bouma, A., Elbers, A.R.W., Dekker, A., De Koeijer, A., Bartels, C., Vellema, P., Van der Wal, P., Van Rooij, E.M.A., Pluimers, F.H. and De Jong, M.C.M. (2003). The food-and-mouth disease epidemic in The Netherlands in 2001. Prev. Vet. Med. 57, 155-166

Greutink, T., Van Klink, E., Van der Peet, G., Mourits, M. and Velthuis, A. (2002). Evaluatie verplaatsingsregelingen MKZ. Expertisecentrum LNV, Ministerie van Landbouw, Natuurbeheer en Visserij. Ede.

Nielen, M., Jalvingh, A.W., Horst, H.S., Dijkhuizen, A.A., Maurice, H., Schut, B.H., Van Wuijckhuise, L.A., and De Jong, M.F. (1996a). Quantification of contacts between Dutch farms to assess the potential risk of foot-and-mouth disease spread. Prev. Vet. Med. 28, 143-158

Nielen, M., Jansen, F.C.M., Van Wuijckhuise, L.A. and Dijkhuizen, A.A. (1996b). Dutch cattle identification and registration (I and R) system: Analysis of its use for controlling an outbreak of foot and mouth disease. Tijdsch. Diergeneesk. 121, 576-581

Webb, C.R. and Sauter Louis, C. (2002). Investigations into the contact structure of the British sheep population. In: Reid, S.W.J. and Menzies, F.D. (Eds.), Proceedings of the Society for Veterinary Epidemiology and Preventive Medicine,Cambridge, 10-20.

Appendix A1. Contact matrix based on animal movements. The total number of animal movements between the different farm types for periods, 2000 and 2002, respectively.

		Da	**Yo**	**Da/Be**	**Be**	**Da/Tr**	**Tr**	**Im/Be**	**Su**	**Cc**	**C-Cc**	**Others**	**Sl**	**New**	**Des**	**Foreign**	**Total**
Da	2000	60420	7751	3888	200575	2659	82783	8876	9009	261268	257919	19432	221076	8	55619	3553	1194836
Da	**2002**	**85808**	**8390**	**4262**	**640303**	**4403**	**166118**	**29355**	**12942**	**6505**	**477573**	**29975**	**486292**	**30628**	**126871**	**21160**	**2130585**
Yo	2000	5704	845	375	751	741	1832	16	134	315	1776	408	2115	0	408	119	15539
Yo	**2002**	**5510**	**462**	**175**	**1060**	**710**	**1549**	**14**	**145**	**16**	**1879**	**485**	**4378**	**193**	**754**	**687**	**18017**
Da/Be	2000	2081	253	467	918	80	1176	95	216	1873	1467	505	7399	1	883	150	17564
Da/Be	**2002**	**2083**	**225**	**167**	**1783**	**85**	**1303**	**82**	**220**	**5**	**2073**	**548**	**10107**	**170**	**1217**	**514**	**20582**
Be	2000	1100	266	278	49454	163	4550	2294	372	2226	1911	828	986148	0	38157	9123	1096870
Be	**2002**	**1614**	**318**	**253**	**24339**	**185**	**4449**	**409**	**377**	**1**	**5287**	**1022**	**1028526**	**2799**	**44810**	**48002**	**1162391**
Da/Tr	2000	1355	562	124	2208	369	1436	158	186	2956	2387	448	3062	0	393	124	15768
Da/Tr	**2002**	**1161**	**474**	**128**	**4746**	**102**	**1533**	**302**	**163**	**10**	**3565**	**366**	**4925**	**232**	**862**	**354**	**18923**
Tr	2000	14927	1543	868	242044	886	27665	13547	2404	22736	19531	5859	60828	14	3867	28664	445383
Tr	**2002**	**12267**	**960**	**572**	**42484**	**940**	**11999**	**3424**	**1396**	**17**	**30994**	**4053**	**68859**	**2430**	**4805**	**25419**	**210619**
Im/Be	2000	208	9	12	276	1	264	175	127	55	28	122	58294	0	2229	98	61898
Im/Be	**2002**	**44**	**7**	**10**	**667**	**1**	**266**	**85**	**29**	**0**	**221**	**63**	**59081**	**62**	**2345**	**2034**	**64915**
Su	2000	3434	149	188	3968	147	3135	255	1848	7369	5231	1807	20429	0	2130	199	50289
Su	**2002**	**4956**	**209**	**448**	**9012**	**192**	**5047**	**613**	**1351**	**29**	**7165**	**2269**	**28931**	**1098**	**3306**	**468**	**65094**
Cc	2000	8654	697	823	193649	970	28415	8551	3882	11749	16231	6372	74357	2	52	21	354425
Cc	**2002**	**18**	**1**	**0**	**2795**	**0**	**106**	**196**	**0**	**0**	**597**	**2**	**8**	**134**	**7**	**9**	**3873**
C-Cc	2000	5115	380	811	280615	448	22547	12898	2257	10950	23303	2800	47297	0	191	31808	441420
C-Cc	**2002**	**203**	**86**	**112**	**67958**	**5**	**9598**	**3941**	**76**	**412**	**24513**	**470**	**13821**	**2604**	**373**	**62860**	**187032**
Others	2000	13475	752	738	5196	515	8990	289	2560	13579	9602	8618	26614	6	3032	491	94457
Others	**2002**	**14959**	**674**	**1081**	**10534**	**593**	**10818**	**620**	**2801**	**39**	**9795**	**7796**	**44824**	**2178**	**4280**	**3024**	**114016**
Sl	2000	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Sl	**2002**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**3546**	**0**	**3546**
New	2000	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
New	**2002**	**4743**	**47**	**232**	**3429**	**52**	**1210**	**732**	**408**	**7**	**1769**	**580**	**27781**	**1596**	**2079**	**3633**	**48298**
Foreign	2000	2318	49	540	154382	59	254779	11738	2189	0	81619	988	3	0	12	1	508677
Foreig	**2002**	**3381**	**309**	**732**	**362682**	**27**	**28426**	**20951**	**2212**	**0**	**182**	**2300**	**122**	**13798**	**24**	**260**	**435406**
Total	2000	118811	13257	9112	1134036	7038	437590	58892	25184	335078	421005	48194	1507622	37	106992	74351	4297126
Total	**2002**	**136747**	**12162**	**8172**	**1171792**	**7295**	**242422**	**60724**	**22120**	**7041**	**565613**	**49929**	**1777655**	**57922**	**195279**	**168424**	**4483297**

Appendix A2. The difference in the number of animal movements between the different farm types between the two periods 2000 and 2002.

	Da	**Yo**	**Da/Be**	**Be**	**Da/Tr**	**Tr**	**Im/Be**	**Su**	**Cc**	**C-cc**	**Others**	**Sl**	**New**	**Des**	**Foreign**	**Total**
Da	5248	-1945	-922	372870	858	55741	17520	930	-341852	133681	4066	191524	30617	16423	52712	537470
Yo	-2095	-665	-325	59	-278	-894	-7	-34	-404	-489	-59	1558	193	528	210	-2702
Da/Be	-692	-112	-456	559	-22	-265	-45	-68	-2492	117	-125	242	169	314	40	-2837
Be	147	-37	-118	-41600	-32	-1618	-2650	-119	-2967	2739	-82	-286338	2799	35838	-6066	-300102
Da/Tr	-646	-275	-37	1802	-390	-382	91	-85	-3931	382	-231	842	232	189	338	-2101
Tr	-7636	-1097	-585	-280241	-241	-24888	-14639	-1809	-30298	4953	-3759	-12245	2411	-12800	-351	-383225
Im/Be	-233	-5	-6	299	0	-86	-148	-140	-73	184	-100	-18644	62	1903	-627	-17616
Su	377	10	197	3721	-4	867	273	-1113	-9796	190	-140	1692	1098	203	466	-1958
Cc	-11521	-928	-1097	-255404	-1293	-37781	-11205	-5176		-21044	-8494	-99135	131	-19	-62	-468694
C-Cc	-6617	-421	-969	-306195	-592	-20465	-13256	-2933	-14188	-6558	-3263	-49242		20449	118	-401528
Others	-3008	-329	97	3606	-94	-1169	235	-612	-18066	-3008	-3695	9339	2170	2369	237	-11927
Sl	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3546	3546
New	4743	47	232	3429	52	1210	732	408	7	1769	580	27781	1596	2079	3633	48298
For	290	244	12	156839	-52	-311279	5300	-707	0	-108643	983	118	0	259	8	-242830
Total	-21641	-5513	-3977	-340256	-2089	-341007	-17799	-11459	-424062	4273	-14320	-232508	41479	67735	54202	-1246204

Appendix B1. Contact matrix based on transport movements. The total number of transports between the different farm types for periods, 2000 and 2002, respectively.

		Da	Yo	Da/Be	Be	Da/Tr	Tr	Im/Be	Su	Cc	C-Cc	Others	Sl	New	Des	Foreign	Total
Da	2000	37785	2802	2162	175956	1456	57534	7966	5506	199241	200986	12062	145570	53123	555	5	902709
Da	**2002**	**39384**	**2454**	**1892**	**494005**	**2138**	**113002**	**23033**	**6474**	**4766**	**351420**	**15934**	**258108**	**118005**	**7177**	**17928**	**1455720**
Yo	2000	2154	210	78	129	212	681	6	63	183	918	186	989	331	33	0	6173
Yo	**2002**	**2220**	**114**	**43**	**320**	**203**	**535**	**11**	**59**	**13**	**827**	**228**	**1604**	**440**	**59**	**49**	**6725**
Da/Be	2000	1151	67	228	563	46	575	39	101	1178	850	245	2855	816	36	1	8751
Da/Be	**2002**	**965**	**62**	**70**	**1207**	**41**	**569**	**66**	**87**	**4**	**1098**	**252**	**3794**	**1073**	**75**	**60**	**9423**
Be	2000	589	46	67	3580	63	656	113	131	1086	775	311	31066	28243	339	0	67065
Be	**2002**	**568**	**50**	**43**	**1845**	**51**	**656**	**74**	**118**	**1**	**809**	**262**	**30395**	**32776**	**1365**	**228**	**69241**
Da/Tr	2000	876	192	52	1790	232	865	131	119	1892	1697	291	1767	371	10	0	10285
Da/Tr	**2002**	**601**	**154**	**32**	**3277**	**72**	**924**	**212**	**111**	**8**	**2275**	**225**	**2225**	**789**	**45**	**137**	**11087**
Tr	2000	7125	310	357	11238	377	8123	818	1011	9123	7538	2656	22094	3171	1217	5	75163
Tr	**2002**	**5274**	**173**	**235**	**6071**	**293**	**4187**	**409**	**604**	**15**	**5364**	**1814**	**22451**	**3953**	**986**	**620**	**52449**
Im/Be	2000	66	2	4	70	1	82	84	35	33	18	52	2128	1415	5	0	3995
Im/Be	**2002**	**21**	**1**	**1**	**67**	**1**	**41**	**23**	**7**	**0**	**37**	**30**	**1924**	**1640**	**62**	**11**	**3866**
Su	2000	2045	82	100	2823	104	1900	211	1074	4934	3381	1094	9683	2022	39	0	29492
Su	**2002**	**2273**	**75**	**175**	**6239**	**121**	**2611**	**433**	**641**	**23**	**4691**	**1112**	**13444**	**3101**	**147**	**495**	**35581**
Cc	2000	5041	310	330	8943	517	7273	387	1864	1213	1641	3643	5583	35	6	2	36788
Cc	**2002**	**6**	**1**	**0**	**82**	**0**	**5**	**5**	**0**	**0**	**30**	**1**	**8**	**6**	**1**	**6**	**151**
C-Cc	2000	2971	117	206	10375	248	3674	678	920	1497	1924	1554	3147	151	1061	0	28523
C-Cc	**2002**	**117**	**8**	**6**	**2704**	**3**	**378**	**126**	**31**	**16**	**955**	**93**	**1371**	**280**	**1733**	**86**	**7907**
Others	2000	7533	288	387	3791	308	5237	248	1497	9185	6469	5359	16683	2831	115	3	59934
Others	**2002**	**7183**	**266**	**424**	**6925**	**342**	**5800**	**476**	**1502**	**30**	**5704**	**4480**	**24763**	**3921**	**316**	**1086**	**63218**
Sl	2000	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Sl	**2002**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**0**	**740**	**0**	**0**	**740**
New	2000	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
New	**2002**	**1264**	**10**	**32**	**1456**	**13**	**529**	**96**	**110**	**5**	**1052**	**264**	**2446**	**1577**	**158**	**242**	**9254**
Foreign	2000	299	7	40	1447	6	3943	457	180	0	528	206	2	0	1	0	7116
Foreig	**2002**	**378**	**13**	**50**	**3826**	**4**	**1427**	**282**	**184**	**0**	**81**	**251**	**27**	**21**	**14**	**193**	**6751**
Total	2000	67644	4434	4011	220705	3570	90550	11138	12501	229567	226725	27663	241567	92527	3417	18	1236037
Total	**2002**	**60254**	**3381**	**3003**	**528024**	**3282**	**130664**	**25246**	**9928**	**4881**	**374343**	**24946**	**362560**	**168322**	**12138**	**21141**	**1732113**

Appendix B2. The difference in the number of transport movements between the different farm types between the two periods 2000 and 2002.

	Da	Yo	Da/Be	Be	Da/Tr	Tr	Im/Be	Su	Cc	C-cc	Others	Sl	New	Des	Foreign	Total
Da	-10996	-1282	-991	259397	197	36290	12412	-867	-260889	83439	-149	64015	17921	47174	6437	252108
Yo	-652	-166	-61	148	-80	-373	3	-25	-231	-397	-20	285	49	-1	15	-1506
Da/Be	-570	-27	-234	456	-20	-198	14	-48	-1567	-35	-75	-13	59	-15	27	-2245
Be	-217	-11	-46	-2928	-33	-219	-77	-57	-1447	-224	-153	-11026	228	-4881	913	-20179
Da/Tr	-567	-102	-37	890	-237	-229	37	-48	-2515	12	-163	-131	137	294	32	-2626
Tr	-4226	-240	-241	-8913	-210	-6644	-682	-744	-12149	-4687	-1727	-7008	613	-275	-637	-47768
Im/Be	-67	-2	-4	-26	0	-68	-89	-40	-44	13	-39	-913	11	-247	55	-1461
Su	-454	-34	42	2475	-18	78	152	-791	-6556	183	-347	533	495	405	95	-3742
Cc	-6715	-412	-440	-11842	-689	-9692	-511	-2485	-1617	-2158	-4856	-7436	3	-41	-7	-48900
C-Cc	-3844	-148	-269	-11129	-328	-4521	-778	-1196	-1980	-1610	-1979	-2825	86	79	318	-30124
Others	-2861	-118	-92	1870	-69	-1183	145	-494	-12217	-2921	-2665	2519	1082	146	163	-16694
Sl	0	0	0	0	0	0	0	0	0	0	0	0	0	740	0	740
New	1252	9	32	1456	13	520	96	110	2	1052	259	2446	239	1553	158	9197
For	-21	4	-3	1897	-4	-3830	-327	-56	0	-623	-24	24	193	21	13	-2737
Total	-29938	-2531	-2345	233751	-1478	9931	10395	-6740	-301208	72043	-11938	40471	21117	44953	7582	84064

RATE AND STRUCTURE OF CATTLE TRANSFERS BETWEEN CATTLE HERDS CONSIDERED TO BE FREE OF PARATUBERCULOSIS

M.F. WEBER[*], H.J.W. VAN ROERMUND, H.B.J. ASSINK AND J.A. STEGEMAN

SUMMARY

The rate and structure of cattle transfers between 206 Dutch cattle herds, with a '*Mycobacterium avium* subsp. *paratuberculosis* (*Map*)-free' status by November 2002, were analysed over a three-year period (November 1999 – November 2002). Of the 206 '*Map*-free' herds, 184 were closed herds. In total, 280 cattle had been introduced into 22 herds at an average rate of 0.33 per year per 100 animals present in the 206 herds. Forty-four of these cattle originated from 12 other '*Map*-free' herds. A further 236 cattle, not originating from a '*Map*-free' herd had been introduced before their herds of destination obtained the '*Map*-free' status. The cattle transfers were not randomly distributed over the herds. No cattle were introduced into the 12 herds from which cattle were transferred to other '*Map*-free' herds. Thus, continued propagation of the infection by cattle transfers was impossible in the group of herds studied during the study period.

INTRODUCTION

Certified *Mycobacterium avium* subsp. *paratuberculosis* (*Map*)-free cattle herds form an important component within a control programme for paratuberculosis (Johne's disease) as they provide a source of non-infected cattle. In The Netherlands, herds can obtain '*Map*-free' status following five annual herd examinations, provided all test results are negative (Benedictus et al., 2000; Weber et al., 2004). Until recently, the status of these herds was subsequently surveyed by annual herd examinations.

It is generally assumed that purchase of cattle is the main route of between-herd transmission of *Map* (Sweeney, 1996). However, to achieve certain goals of the farmer, such as optimising production, purchase of cattle may be necessary. To reduce the risk of introduction of a *Map* infection, the Dutch paratuberculosis certification-and-surveillance programme requires that cattle may be added into a '*Map*-free' herd only if the cattle originate from another '*Map*-free' herd. In addition, cattle may be added to herds that are in the process of '*Map*-free' certification only if they originate from a herd with an equal or higher number of negative annual herd examinations. However, cattle that have been raised in a '*Map*-free' herd are allowed to return to this herd after a period of shared grazing in a herd with a lower status, because the risk of introducing *Map* through such re-introductions is currently considered small.

---

[*] Maarten F. Weber, Animal Health Service, P.O. Box 9, 7400 AA Deventer, The Netherlands.
E-mail: m.weber@gdvdieren.nl

At present, there are approximately 24,000 dairy herds in The Netherlands. In May 2000, the first Dutch dairy herd obtained the '*Map*-free' status and, at 1st December, 2003, there were 352 '*Map*-free' herds in The Netherlands. However, the surveillance of '*Map*-free' herds by annual herd-examinations was considered expensive and an effective but cheaper surveillance programme was required. To maintain a group of herds free of *Map* long-term, an infected herd should be detected by the surveillance programme and removed from the group before the infection has spread to, on average, one other herd. Then, only small outbreaks would be expected if a pre-existing infection remained undetected in a certified '*Map*-free' herd or if there was inadvertent introduction of *Map* into a certified '*Map*-free' herd. Transmission between herds is determined by the probability of infection given contact, the contact rate and the contact structure (that is, which herds are in contact with other herds). To study alternative test schemes in a surveillance programme, an analytical model for between-herd transmission was developed (van Roermund et al., 2002). Between-herd transmission was found to be linearly dependent on the contact rate if a random contact structure was assumed, while the slope of this relationship varied between alternative test schemes. Thus, knowledge of the rate and structure of contacts between '*Map*-free' herds is pivotal for the design of an effective surveillance programme. Therefore, in the present study, the rate and structure of cattle transfers between '*Map*-free' herds were studied.

MATERIALS AND METHODS

Study population and data collection

All 206 Dutch cattle herds with a '*Map*-free' status on 1st November 2002 were selected from the national cattle identification and registration (I&R) system (Nielen et al., 1996). For each selected herd, data on all cattle present in the herd, introduced into the herd, moved to another herd, exported to other countries or that had died or were culled between 1st November 1999 and 1st November 2002 were retrieved from the I&R system. Furthermore, the herd type (dairy, beef or other) and the date at which the '*Map*-free' status was obtained were retrieved for each herd. For each animal that was transferred to a selected herd or transferred from a selected herd to another herd, the identification of the herd of origin or destination was retrieved. For each herd of origin or destination, the paratuberculosis status at 1st November 2002 was retrieved.

Quality control of data

The data sets were extensively checked for inconsistencies. Furthermore, consistency of the data in relation to the regulations of the paratuberculosis certification and surveillance programme was also checked.

Data analysis

For each selected herd with a '*Map*-free' status on 1st November 2002, the average number of animals present in each herd in the first year of the study (1st November 1999 to 1st November 2000), the second year of study (1st November 2000 to 1st November 2001) and the third year of study (1st November 2001 to 1st November 2002) was approximated by the mean of the number present at the beginning and the end of each year. All recorded cattle transfers were included in a matrix of the herds of origin and destination. For each selected herd with herd number 'h', the number of animals introduced (I) into the herd from another herd in year 'y' was expressed as the proportion $I_{h,y}$ of the estimated average number of animals present in that year in the herd concerned:

$$I_{h,y} = \frac{number\ of\ animals\ introduced_{(h,y)}}{estimated\ average\ number\ of\ animals_{(h,y)}}$$

Similarly, the annual proportion of animals moved ($M_{h,y}$) from each of the selected herds to another herd was expressed as:

$$M_{h,y} = \frac{number\ of\ animals\ moved\ to\ another\ herd_{(h,y)}}{estimated\ average\ number\ of\ animals_{(h,y)}}$$

Then, the average annual rate of animal introduction ($\overline{\overline{I}}$) and movement ($\overline{\overline{M}}$) from the 206 selected herds during the three years of study were calculated as

$$\overline{\overline{I}} = \frac{1}{206} \cdot \frac{1}{3} \cdot \left(\sum_{h=1}^{h=206} \sum_{y=1}^{y=3} I_{h,y} \right) yr^{-1} \quad \text{and}$$

$$\overline{\overline{M}} = \frac{1}{206} \cdot \frac{1}{3} \cdot \left(\sum_{h=1}^{h=206} \sum_{y=1}^{y=3} M_{h,y} \right) yr^{-1} \quad \text{respectively.}$$

Equality of the median proportion of animals introduced into the selected '*Map*-free' herds in year 1, 2 and 3 was tested with the Friedman test by year blocked for herd, using the statistical computer package, Minitab® release 12.1 (Minitab Ltd, Coventry, U.K.). Similarly, equality of the median proportion of animals moved from the selected '*Map*-free' herds to other herds in year 1, 2 and 3 was tested. Statistical significance was declared at $p \leq 0.05$.

RESULTS

Of the 206 selected herds, 194 herds were registered in the I&R system as a dairy herd, 8 herds were registered as a mixed dairy and beef herd, and 4 herds were registered as a beef or other herd. These herds are indicated as '*Map*-free' herds throughout this paper, although they had actually obtained this status at a varying time between early May 2000 and late October 2002 (Fig. 1). The location of these herds in The Netherlands is shown in Fig. 2. The mean herd size (±SD) of the 206 herds was 115 (±52) cattle, including young stock (Fig. 3).

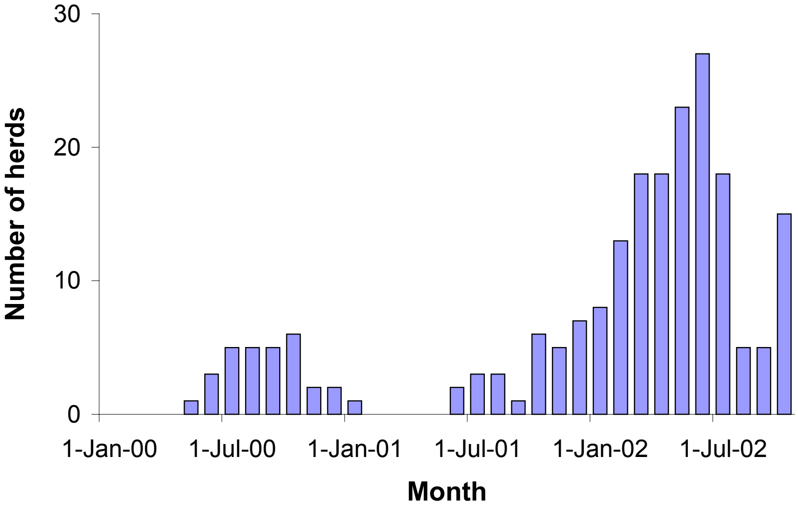


Fig. 1. Time at which the 206 herds studied obtained their '*Map*-free' status.

Fig. 2. Distribution over The Netherlands of all 206 Dutch herds with a '*Map*-free' status on 1st November 2002.

Introduction of cattle into '*Map*-free' herds

In all, 280 cattle were introduced into 22 (11%) of the 206 herds (Fig. 4). As 223 of these 280 animals were introduced into only four of these 22 herds (Fig 5A), the distribution of the annual rate of introduction over this three-year period for these 22 herds was positively skewed (Fig. 5B). For the majority of cases (19 of the 22 herds), cattle were introduced from only one herd each whilst only one herd received cattle from two herds, another herd introduced cattle from three herds and one herd received cattle from five different herds. In the remaining 184 herds (89%), no cattle were introduced from another herd during the three years studied. The overall average rate of introduction $\bar{\bar{I}}$ in the 206 herds was 0.33 per year per 100 animals present. There was no significant difference in the median rate of introduction between the three years (S=0.09, df=2, p=0.956). Forty-four of the 280 transferred cattle originated from another herd with a '*Map*-free' status on 1st November 2002. Eighty-four cattle originated from a herd that was in the process of certification at the time of transfer, but had not yet obtained the '*Map*-free' status at 1st November 2002. Hundred-and-forty-six transferred cattle were raised until at least 6 months old in a '*Map*-free' herd and returned to this same herd after a period of shared grazing elsewhere. The remaining six cattle were transferred from herds with an unknown paratuberculosis status to '*Map*-free' herds, in conflict with the regulations of the paratuberculosis certification and surveillance programme. Upon request of the Animal Health Service, these six cattle were subsequently removed from those herds, after which the herds regained their '*Map*-free' status.

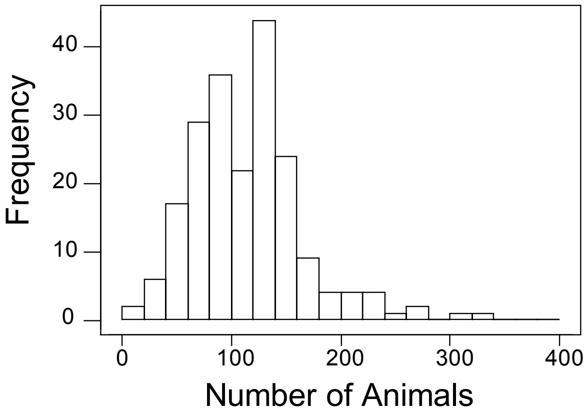


Fig. 3. Histogram of herd size, including young stock, of cattle (n=23560) in 206 herds with a '*Map*-free' status on 1st November 2002.

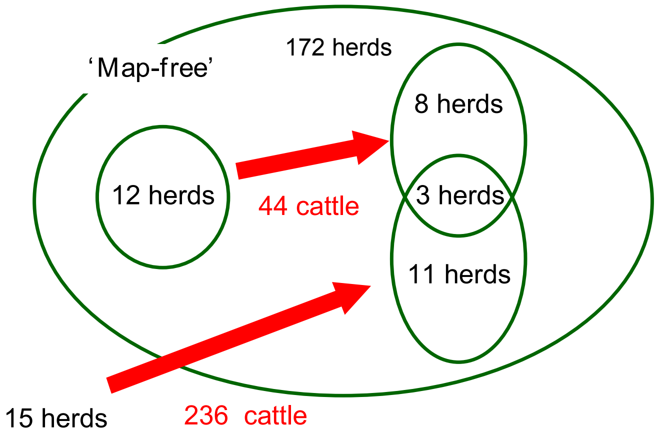


Fig. 4. Cattle introductions observed during a three-year period in 206 '*Map*-free' herds.

Cattle transferred from ‘*Map*-free’ herds

During the three years, 24526 cattle, including calves for veal production, were transferred from the 206 ‘*Map*-free’ herds to other herds. Of these, 44 cattle from 12 herds were transferred to 11 other ‘*Map*-free’ herds (Fig. 4). No cattle were introduced into any of the 12 ‘*Map*-free’ herds of origin during the period of study. The distribution of the average annual rate of cattle moved from these 12 herds was positively skewed (Fig. 6). For all 206 ‘*Map*-free’ herds, the average rate $\overline{\overline{M}}$ of cattle transferred to other ‘*Map*-free’ herds during the three years of study equalled 0.6×10^{-3} yr^{-1}. For all 206 ‘*Map*-free’ herds, the average rate $\overline{\overline{M}}$ of cattle transferred to herds still in the process of obtaining this status on 1st November 2002 equalled 3.1×10^{-3} yr^{-1}. The median rates of cattle transferred from ‘*Map*-free’ herds to other ‘*Map*-free’ herds in year 1, 2 and 3 were not significantly different (S=0.42, df=2, p=0.809). Neither were the median rates of cattle transferred from ‘*Map*-free’ herds to herds that where in the process of obtaining a ‘*Map*-free’ status on 1st November 2002 (S=2.67, df=2, p= 0.263).

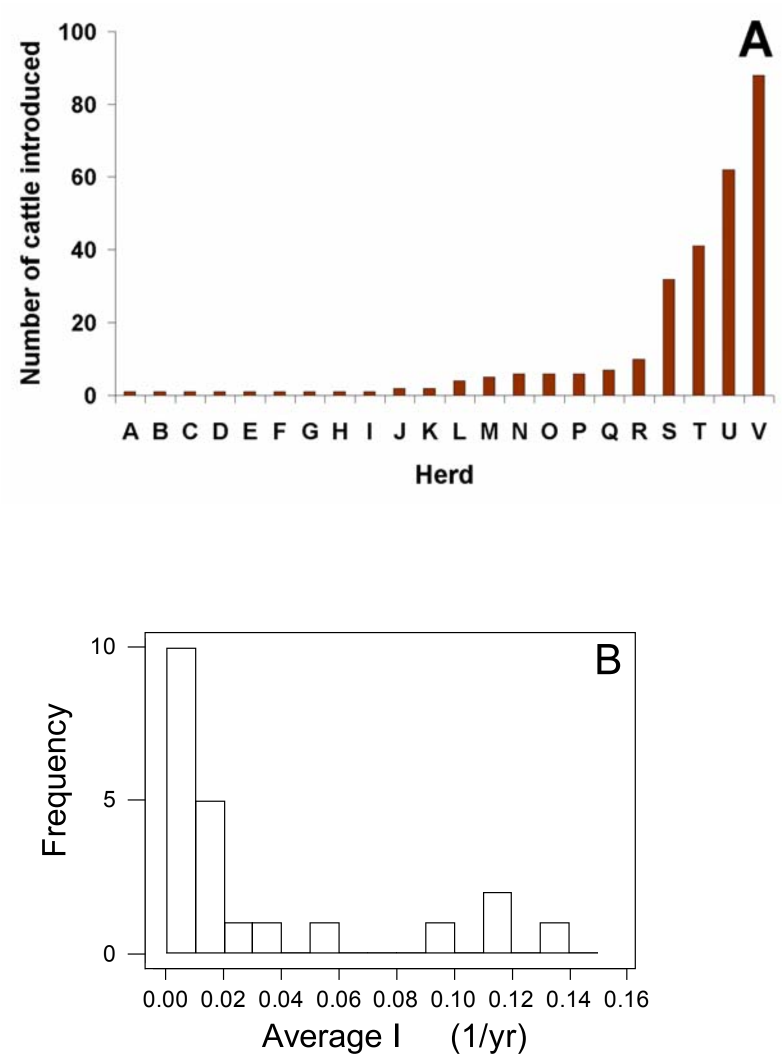


Fig 5. Number of cattle introduced over a three-year period (A) and average rate of introduction over a three-year period (B) for the 22 ‘*Map*-free’ herds into which cattle were introduced.

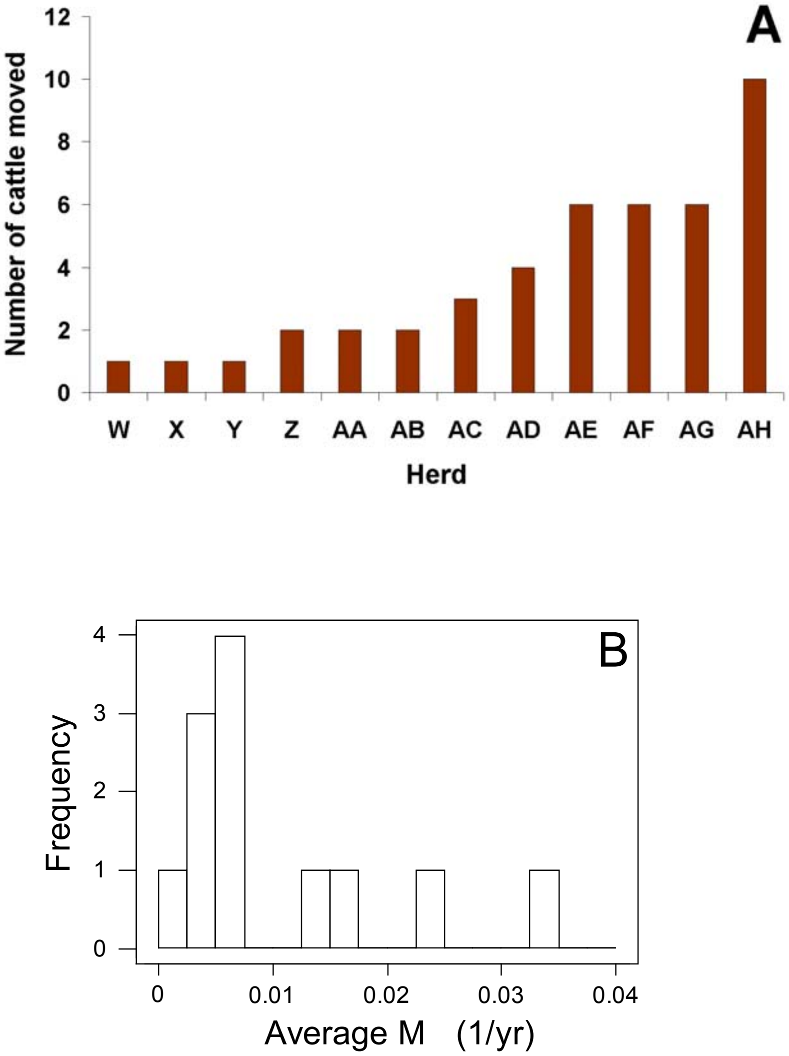


Fig. 6. Number of cattle moved to other 'Map-free' herds over a three-year period (A) and average rate of movement of cattle to '*Map*-free' herds over a three-year period (B) for 12 '*Map*-free' herds of origin.

DISCUSSION

Limitation of potential between-herd transmission is a key element of a successful surveillance programme for herds considered to be free of an infectious disease. The rate and structure of cattle transfers between herds are major determinants of the between-herd transmission of *Map*. In the present study, the rate and structure of cattle transfers between Dutch '*Map*-free' herds were characterised to enable the design of a cost-effective alternative to the Dutch surveillance programme for '*Map*-free' herds.

The results of the present study show that the majority of the '*Map*-free' herds were closed. The observed rates of cattle transfers between '*Map*-free' herds were lower than the observed rates of transfers in the general Dutch dairy population of 0.02 to $0.03 \cdot yr^{-1}$ per animal present (Graat et al., 2001; Velthuis, 2004). Possibly, owners of certified '*Map*-free' herds are more aware of risks associated with purchase of cattle then other farmers and they are therefore less likely to introduce purchased cattle into their herds. Furthermore, the observed contact structure was not random. No cattle were transferred to herds that were themselves a source of cattle for other '*Map*-free' herds. Thus, continued propagation of *Map*-infection by cattle transfer was impossible in this group of herds during the three years observed in this study.

Until recently, '*Map*-free' herds in the current Dutch paratuberculosis programme were monitored by annual faecal herd examination consisting of a pooled faecal culture of all cattle over 2 years of age (Kalis et al., 2000). However, at the rates of transfer of cattle observed in the present study, biennial herd examinations were also expected to result in a sufficiently low between-herd transmission rate if a random contact structure was assumed (van Roermund et al., 2002). Therefore, a surveillance scheme was recently adopted in which closed '*Map*-free' herds are monitored by biennial faecal herd examinations by pooled faecal culture of all cattle over 2 years. To increase awareness amongst farmers of the risks associated with the introduction of cattle, '*Map*-free' herds in which cattle were introduced during the previous year are still monitored by annual faecal herd examination. However, the assumption of a random contact structure in the study by van Roermund et al. (2002) is in contrast with our finding of a non-random contact structure in which no cattle were transferred to herds that were themselves a source of cattle for other '*Map*-free' herds. Given a non-random contact structure, the surveillance programme may possibly be further relaxed. Further modelling studies of surveillance programmes for infectious diseases in which a non-random contact structure is incorporated are required. An effective surveillance scheme might then be differentiated with respect to the risk posed by transfer of cattle from '*Map*-free' herds to other '*Map*-free' herds and to herds with a lower status.

To gain insight in the medium-term structure of transfers between herds, all cattle transfers to and from herds with a '*Map*-free' status on 1st November 2002 during the previous three years were included in the present study, even if the herd had not yet obtained the '*Map*-free' status by the time of the transfer (Fig. 1). All '*Map*-free' herds had obtained this status at a point in time during the study period. Obtaining this '*Map*-free' status might influence the contact structure of the herd, for instance if other farmers would prefer to buy cattle from a '*Map*-free' herd. However, no difference was found in the median rates of introduction and movement of cattle between the first, second and third year of study. Therefore, the data from the three years of study were pooled, irrespective of the time at which a herd had obtained the '*Map*-free' status.

The majority of cattle introduced into herds with a '*Map*-free' status on 1st November 2002 originated from a herd without this status by that time. At first sight, this may seem in conflict with the regulation that herds are only allowed to introduce cattle from herds with an equal or higher paratuberculosis status. However, approximately half of the cattle transferred to a '*Map*-free' herd were in fact raised until at least six months old in this '*Map*-free' herd and returned to this same herd after a period of shared grazing elsewhere. Another 30 percent of cattle transferred to a herd with a '*Map*-free' status on 1st November 2002 originated from a herd that was in the process of obtaining this status or the herd was no longer in existence on that date. At the time of transfer, these herds of origin had an equal or higher number of negative annual herd examinations than the herd of destination.

For practical reasons, our study was limited to a three-year period. Over a longer period, the contact structure might alter. For instance, '*Map*-free' herds in which cattle have been introduced in the past, may possibly become a source of cattle for other '*Map*-free' herds in the future. Therefore, continuous monitoring of the contact rate and structure of herds participating in any surveillance programme for an infectious disease is required. Furthermore, contacts between herds considered to be '*Map*-free', other than by transfer of cattle, were assumed to be uncommon and of minor importance because of the geographical distribution of these herds in the present study (Fig. 2). Almost without exception, introduction of paratuberculosis into a susceptible herd is through addition of infected carrier animals (Sweeney, 1996). However, contacts other than by transfer of cattle should be the focus of further studies.

At the time of the present study, the whole target population of Dutch '*Map*-free' herds was included in the study. Therefore, a point-estimate of the average transfer rates $\overline{\overline{M}}$ and $\overline{\overline{I}}$ was calculated and confidence intervals were omitted.

The present study was performed to support the design of a suitable surveillance scheme for Dutch '*Map*-free' herds and it was focussed on the contact structure of these herds alone. Therefore, our results can not be easily extrapolated to other countries or surveillance schemes for other infectious diseases. However, the mechanisms of transmission of *Map* are comparable in other countries. Furthermore, the contact structure between herds is equally important for the between-herd transmission of other infectious diseases. Therefore, the concepts of this study are equally applicable to surveillance schemes for other infectious diseases and to other countries.

In conclusion, the majority of '*Map*-free' herds were closed and the observed rate of cattle transfers between certified '*Map*-free' herds was low. The structure of these cattle transfers was not random. Such characteristics should be incorporated in future model studies on surveillance programmes for paratuberculosis and other infectious diseases.

ACKNOWLEDGEMENTS

The authors would like to thank J. Verhoeff and A. Kummeling for comments on a previous version of the manuscript.

REFERENCES

Benedictus, G., Verhoeff, J., Schukken, Y.H. and Hesselink, J.W. (2000). Dutch paratuberculosis programme history, principles and development. Vet. Microbiol. 77, 399-413

Graat, E.A.M., van Oijen, M.A.A.J., Frankena, K., Boender, G.J., Franken, P. and de Jong, M.C.M. (2001). Design of surveillance programmes: an example with mathematical modelling. Proceedings of the Society of Veterinary Epidemiology and Preventive Medicine, Noordwijkerhout, 28-30 March 2001, 29-36.

Kalis, C.H., Hesselink, J.W., Barkema, H.W. and Collins, M.T. (2000). Culture of strategically pooled bovine fecal samples as a method to screen herds for paratuberculosis. J. Vet. Diagn. Invest. 12, 547-551

Nielen, M., Jansen, F.C., van Wuijckhuise, L.A. and Dijkhuizen, A.A. (1996). [I&R (identification and registration) system cattle: an analysis of its use during a foot-and-mouth-disease outbreak in The Netherlands]. Tijdschr. Diergeneeskd. 121, 576-581

Sweeney, R.W. (1996). Transmission of paratuberculosis. Vet. Clin. North Am. Food. Anim. Pract. 12, 305-312

Van Roermund, H.J.W., Weber, M.F., Graat, E.A.M. and de Jong, M.C.M. (2002). Monitoring programmes for paratuberculosis-unsuspected cattle herds, based on quantification of between-herd transmission. Proceedings of the Seventh International Colloquium on Paratuberculosis, Bilbao, Spain, June 11-14, 2002. Pp. 371-375.

Velthuis, A.G.J. (2004). Effect of regulations on the contact structure of the Dutch cattle sector. Proceedings of the Society of Veterinary Epidemiology and Preventive Medicine, Martigny, Switzerland, 200-215.

Vonk Noordegraaf, A., Nielen, M., Franken, P. and Dijkhuizen, A.A. (2002). Simulation modelling of BHV1 control programme at national level, with special attention to sensitivity analysis. In: Vonk Noordegraaf, A. Simulation modelling to support national policy making in the control of bovine herpesvirus 1. PhD thesis, Wageningen University. Pp. 63-89.

Weber, M.F., Groenendaal, H., van Roermund, H.J.W. and Nielen, M. (2004). Simulation of alternatives for the Dutch Johne's disease certification and monitoring program. Prev. Vet. Med. 62, 1-17.

Society for Veterinary Epidemiology and Preventive Medicine

PAST PRESIDENTS

1982-’83	G. Davies
1983-’84	P.R. Ellis
1984-’85	G. Gettinby
1985-’86	R.W.J. Plenderleith
1986-’87	M.V. Thrusfield
1987-’88	K.S. Howe
1988-’89	A.M. Russell
1989-’90	S.G. McIlroy
1990-’91	J.E.T. Jones
1991-’92	J.M. Booth
1992-’93	E.A. Goodall
1993-’94	R.G. Eddy
1994-’95	J.T. Done
1995-’96	G.J. Gunn
1996-’97	M.S. Richards
1997-’98	J.D. Collins
1998-’99	F.D. Menzies
1999-’00	K.L. Morgan
2000-’01	S.W.J. Reid
2001-’02	A.D. Paterson
2002-’03	L.E. Green

EXECUTIVE COMMITTEE 2003-2004

J.L.N. Wood (President), L.E. Green (Senior Vice-President), E.G.M. van Klink (Junior Vice-President), J.R. Newton (Honorary Secretary), D.J. Mellor (Honorary Treasurer), N. Honhold, L. Kelly, E.J. Peeler, K.D.C. Stärk, S.W.J. Reid (Proceedings Editor), F.D. Menzies (Proceedings Editor), M.G. Doherr (co-opted), G.J. Gunn (co-opted)

Honorary Auditors: J.M. Booth, R.G. Eddy

LIFE MEMBERS

J.M. Booth, M.J. Clarkson, G. Davies, R.G. Eddy, P.R. Ellis,
E.A. Goodall, A.M. Russell, M.V. Thrusfield

PLENARY TALKS

Year	Gareth Davies Lecture	Conference Opening Plenary
2004	Ulrich Kihm: BSE and the stable to table concept	Gary Smith: Spatial models of infectious disease in the USA: a crisis of conference and confidentiality
2003	Sir David Cox: The current state of statistical science	Ynte Schukken: Molecular and mathematical epidemiology of bovine mastitis
2002	George Gettinby: Informatics and epidemiology – the first 400 years	Bryan Grenfell: Deterministic and stochastic influences on the dynamics and control of infectious diseases
2001	Will Houston: Science politics and animal health policy: epidemiology in action	Mart de Jong: Design and analysis of transmission experiments
2000	Jim Scudamore: Surveillance – past, present and future	Dirk Pfeiffer: Spatial analysis – a new challenge for veterinary epidemiologists
1999	Aalt Dijkhuizen: The 1997/98 outbreak of classical swine fever in the Netherlands: lessons learned from an economic perspective	Mark Woolhouse: Understanding the epidemiology of scrapie
1998	Wayne Martin: Art, science and mathematics revisited: the role of epidemiology in promoting animal health	-

SOCIETY FOR VETERINARY EPIDEMIOLOGY AND PREVENTIVE MEDICINE

APPLICATION FOR MEMBERSHIP

Name ....

Address ....

....

....

....

Telephone: ....

Fax: ....

E-mail:...

Signed .... Date ....

Please enclose the membership fee (£20 sterling) along with this application form. Overseas members without British bank accounts are requested to pay 2 - 3 years in advance. Cheques should be in £ sterling and drawn from a British bank. British members should pay future dues by standing order (forms are available from the Secretary or Treasurer). Payment can also be made by credit card; the appropriate form is available from the Society's web site, http://www.svepm.org.uk/, or from the Secretary or Treasurer.

Please send this form to the Society's Treasurer:

Dr Dominic Mellor
Comparative Epidemiology and Informatics
Department of Veterinary Clinical Studies
University of Glasgow Veterinary School
Bearsden Road
Glasgow
G61 1QH

☎ +44 (0) 141 330 5742
FAX +44 (0) 141 330 5729
Email d.mellor@vet.gla.ac.uk

Please turn over

INTEREST GROUPS

Please tick appropriate boxes to indicate your interests:

- ☐ Analytical Epidemiology (Observational Studies)
- ☐ Quantitative Epidemiology & Statistical Techniques (Incl. Statistical/Mathematical Modelling)
- ☐ Herd/Flock Level Disease Control Strategies
- ☐ National/International Disease Control Policy
- ☐ Sero-Epidemiology
- ☐ Herd Health and Productivity Systems
- ☐ Disease Nomenclature and Epidemiological Terminology
- ☐ Economic Effects of Disease on Animal Production
- ☐ Veterinary Public Health and Food Hygiene
- ☐ Computing, including data logging
- ☐ Computer Programming *per se*
- ☐ Population and Animal Disease Databases
- ☐ Information System Design
- ☐ Geographical Information Systems (GIS)
- ☐ Risk Analysis

CONSTITUTION AND RULES

NAME

1. The society will be named the Society for Veterinary Epidemiology and Preventive Medicine.

OBJECTS

2. The objects of the Society will be to promote veterinary epidemiology and preventive medicine.

MEMBERSHIP

3. Membership will be open to persons either actively engaged or interested in veterinary epidemiology and preventive medicine.
4. Membership is conditional on the return to the Secretary of a completed application form and subscription equivalent to the rate for one calendar year. Subsequent subscriptions fall due on the first day of May each year.
5. Non-payment of subscription for six months will be interpreted as resignation from the Society.

OFFICERS OF THE SOCIETY

6. The Officers of the Society will be President, Senior Vice-President, Junior Vice-President, Honorary Secretary and Honorary Treasurer. Officers will be elected annually at the Annual General Meeting, with the exception of the President and Senior Vice-President who will assume office. No officer can continue in the same office for longer than six years.

COMMITTEE

7. The Executive Committee of the Society normally will comprise the officers of the Society and not more than four ordinary elected members. However, the Committee will have powers of co-option.

ELECTION

8. The election of office bearers and ordinary committee members will take place at the Annual General Meeting. Ordinary members of the Executive Committee will be elected for a period of three years. Retiring members of the Executive Committee will be eligible for re-election. Members will receive nomination forms with notification of the Annual General Meeting. Completed nomination forms, including the signatures of a proposer, seconder, and the nominee, will be returned to the Secretary at least 21 days before the date of the Annual General Meeting. Unless a nomination is unopposed, election will be by secret ballot at the Annual General Meeting. Only in the event of there being no nomination for any vacant post will the Chairman take nominations at the Annual General Meeting. Tellers will be appointed by unanimous agreement of the Annual General Meeting.

FINANCE

9. An annual subscription will be paid by each member in advance on the first day of May each year. The amount will be decided at the annual general meeting and will be decided by a simple majority vote of members present at the Annual General Meeting.

10. The Honorary Treasurer will receive, for the use of the Society, all monies payable to it and from such monies will pay all sums payable by the Society. He will keep account of all such receipts and payments in a manner directed by the Executive Committee. All monies received by the Society will be paid into such a bank as may be decided by the Executive Committee of the Society and in the name of the Society. All cheques will be signed by either the Honorary Treasurer or the Honorary Secretary.
11. Two auditors will be appointed annually by members at the Annual General Meeting. The audited accounts and balance sheet will be circulated to members with the notice concerning the Annual General Meeting and will be presented to the meeting.

MEETINGS

12. Ordinary general meetings of the Society will be held at such a time as the Executive Committee may decide on the recommendations of members. The Annual General Meeting will be held in conjunction with an ordinary general meeting.

GUESTS

13. Members may invite non-members to ordinary general meetings.

PUBLICATION

14. The proceedings of the meetings of the Society will not be reported either in part or in whole without the written permission of the Executive Committee.
15. The Society may produce publications at the discretion of the Executive Committee.

GENERAL

16. All meetings will be convened by notice at least 21 days before the meeting.
17. The President will preside at all general and executive meetings or, in his absence, the Senior Vice-President or, in his absence, the Junior Vice-President or, in his absence, the Honorary Secretary or, in his absence, the Honorary Treasurer. Failing any of these, the members present will elect one of their number to preside as Chairman.
18. The conduct of all business transacted will be under the control of the Chairman, to whom all remarks must be addressed and whose ruling on a point of order, or on the admissibility of an explanation, will be final and will not be open to discussion at the meeting at which it is delivered. However, this rule will not preclude any member from raising any question upon the ruling of the chair by notice of motion.
19. In case of an equal division of votes, the Chairman of the meeting will have a second and casting vote.
20. All members on election will be supplied with a copy of this constitution.
21. No alteration will be made to these rules except by a two-thirds majority of those members voting at an annual general meeting of the Society, and then only if notice of intention to alter the constitution concerned will have appeared in the notice convening the meeting. A quorum will constitute twenty per cent of members.
22. Any matter not provided for in this constitution will be dealt with at the discretion of the Executive Committee.

April, 1982
Revised March, 1985; April, 1988; November 1994
Corrected January 1997; April 2002