

# Effects of different pH on cholinesterases activity and bimolecular rate constant of dichlorvos in the tissues of food animals



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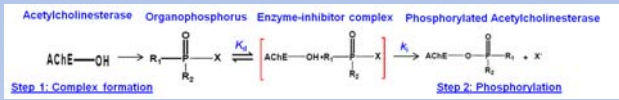
## Background

Cholinesterases (ChEs) are specialized carboxylic ester hydrolases that catalyse the hydrolysis of choline esters. Two types of ChE activity have been identified in mammalian tissues; these are distinguished according to their substrate specificity and sensitivity to the selective inhibitors. The first is acetylcholinesterase (AChE, EC 3.1.1.7). The second is butyrylcholinesterase (BChE, EC 3.1.1.8).

Organophosphorus (OP) compounds are esters of phosphoric acid containing alkoxy, alkyl, amino or thioalkyl groups. These pesticides are widely used today in veterinary medicine to control parasites [1].

The toxic effects of OP [dichlorvos (DDVP)] compounds are almost entirely due to covalent modification and hence inhibition of acetylcholinesterase (AChE) in the nervous system.

An outline of the mechanism of inhibition of AChE by organophosphorus compounds is shown below.



## Objectives

To investigate the optimum pH for AChE and BChE from liver and muscle of sheep, cattle, and pig.

A further aim was to study the most sensitive pH effect to the rate of constants ( $k_i$ ) of DDVP-inhibited AChE and BChE from liver of food animals.

## Methods

Meat from food animals (5 sheep, 5 cattle and 5 pigs) was obtained from local abattoirs and transported in a cool box to the laboratory.

To extract ChEs, samples of liver and muscle were cut into small pieces (3-5 mm<sup>3</sup>), and homogenized using a mechanically-driven homogenizer (Model X520-D, T6 probe, Bennett and Company, Weston-super-Mare, UK) with phosphate buffer, pH 8.0 (ratio 1:9), and centrifuged at 9000 g for 5 min.

AChE activity was determined using the Ellman (1961) method, adapted for a plate reader.

For detection of optimum pHs, ChEs was measured for different pHs (6-9) at 20 °C. The increase and decrease in ChE activity over different pHs was then monitored (e.g. Figure 1A).

For measurement of rate constants for inhibition ( $k_i$ ), ChE was inhibited with 4, 6, and 8 μM DDVP in pHs 6.5, 7.5, and 8.5, respectively. The decrease in ChE activity over different times (0-60 min) was then plotted (e.g. Figure 1B).

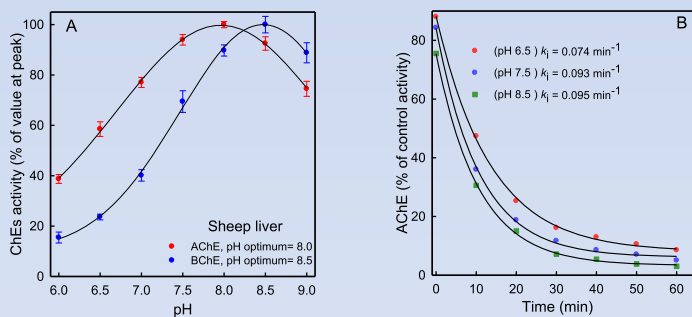


Figure 1: The figures above illustrate the data analysis used to obtain for optimum pH (A) and for rate constant for inhibition (B). The data are for sheep liver. Optimum pH data were fitted with a Gaussian peak using SigmaPlot 11 (Systat software, Inc.). For the rate constants ( $k_i$ ) data the inhibition time courses at different times after inhibition were fitted with a single exponential decay.

## Acknowledgements

This research was supported by a Ph.D. studentship from the Iraqi Ministry of Higher Education.

## Results

Table 1: Optimum pH values for AChE and BChE from liver and muscle of food animals.

Tissues	Animals	AChE	BChE
Liver	Sheep	8.0	8.5
	Cattle	8.1	8.3
	Pig	7.9	8.5
Muscle	Sheep	8.1	7.8
	Cattle	8.2	7.8
	Pig	8.1	7.7

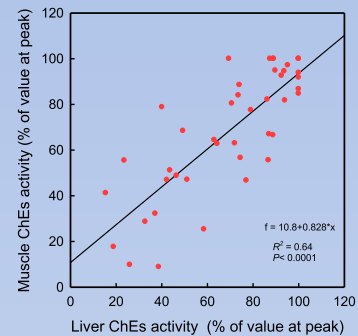


Figure 2: Comparison between liver and muscle ChEs activities (percent of value at peak) from food animals within a range of pHs (6-9).

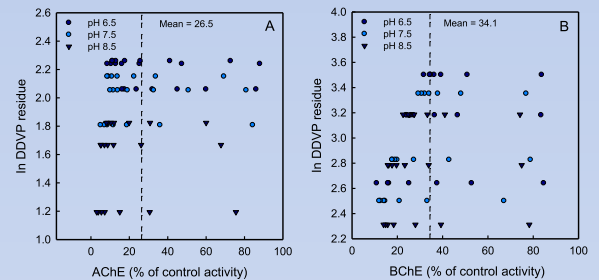


Figure 3: Residues of the DDVP (plotted on the y-axis) vs. the percent liver ChEs activities (x-axis) for food animal at different pHs. Vertical dashed lines are drawn at the mean percent control of ChEs activities.

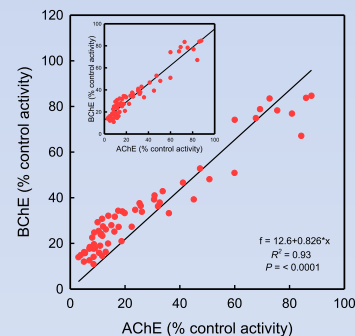


Figure 4: Comparison between AChE and BChE activities (percent control) in liver from sheep, cattle, and pigs inhibited with DDVP within three different pHs (6.5, 7.5, and 8.5).

## Conclusions

- This is the first study that provided original data concerning an enzymological characterization of this inhibitor in food animals.
- In liver, the Optimum pH values in BChE was higher than AChE, while in muscle optimum pH values in AChE higher than BChE.
- In both AChE and BChE,  $k_i$  values for inhibition by DDVP compounds was higher in pH 6.5 than other pHs.
- In all animals residual ChEs activity after inhibition by DDVP compounds was lower in AChE than in BChE.

# Essential Requirements for Surveillance Systems for Emerging Diseases

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## Background

Surveillance systems should support the ongoing systematic collection and analysis of data, resulting in relevant intelligence at an appropriate geographical and temporal scale to support risk managers in taking decisions to prevent and control an emerging disease. Two recent emerging disease events, reported colony losses in bees and the Q fever outbreak in the Netherlands, have highlighted important requirements for existing surveillance systems to ensure preparedness for emerging diseases at EU level.

## Method

For each disease event existing surveillance data for the European Economic Area countries was collated and reviewed. In addition questionnaires were completed by reporting organisations in these countries describing the existing surveillance systems.

## Results-Colony losses in bees

For the 24 countries completing the surveillance network analysis tool (SNAT) a general weakness in most of the surveillance systems was identified (Figure 1). Key system components missing included, technical committee to develop procedures, integration with laboratory services, protocols suitable to collect representative figures, relational data management tools and performance indicators.

The review of the existing surveillance data indicated a lack of representative data at country level and comparable data at EU level for colony losses (Figure 2). The major problems were inconsistent definitions of "colony losses" and the variability and validity of the epidemiological indicators reported.

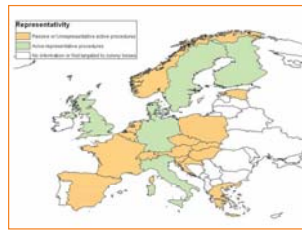


Figure 2. Countries with representative surveillance data in Europe

## Results-Q Fever in ruminants

The responses from 26 countries completing the questionnaire indicated that the disease is notifiable in 14 countries but no harmonised case definition was available. In addition only a few countries operate official surveillance programmes and 5 countries do not have a national reference laboratory established.

The review of available surveillance data highlighted the considerable differences in testing protocols and programme design (Table 1). Consequently interpretation of the data was hampered by missing data, and the inability to discriminate between active and passive systems and prevalence and incidence epidemiological indicators.

Programme	Sampling stage	Test	Number Countries Reporting	Animals Tested	Herds Tested
Clinical investigations	Not reported	CF	1	1	1
Clinical investigations	at autopsy blood	ELISA	1	3	1
Clinical investigations	at farm-blood	CF	2	150	0
Clinical investigations	at farm-blood	ELISA	1	5	1
Clinical investigations	at farm-blood	IFA	1	131	4
Clinical investigations	at farm-organ/tissue	CF	1	19	15
Clinical investigations	at farm-organ/tissue	PCR	1	37	0
Clinical investigations	at farm-organ/tissue	IHC	1	2	0
Surveillance survey	Not reported	CF	1	774	51
Surveillance survey	at farm-blood	ELISA	1	31	1
Surveillance survey	at farm-bulk milk	ELISA	1	0	348
Surveillance-selective sampling	at farm-milk	PCR	1	0	2
Survey	at farm-blood	ELISA	1	513	0
random survey, non ap-parent Q fever	blood	ELISA	1	402	10
random survey, non ap-parent Q fever	vaginal swab	PCR	1	149	10

Table 1. Available data for Q fever testing in Goats for 2009

(CF=complement fixation, PCR=polymerase chain reaction, ELISA=enzyme linked immunosorbent assay, IFA=immunofluorescence assay, IHC=immunohistochemical)



Figure 1. Percentage of surveillance systems for colony losses in bees complying with the 40 SNAT evaluation criteria

## Conclusions

- > Clear and specific case definitions should be specified for all disease events monitored by a system
- > Integration with laboratory services and use of appropriate testing methods is essential, for emerging diseases negative results represent valuable information
- > Consistent and robust epidemiological indicators calculated according to standard protocols for comparable populations should be defined
- > Development of generic data models to facilitate data transfer and analysis at country and EU level is recommended
- > Development of common performance indicators for surveillance systems would result in a robust standardised surveillance at EU level for emerging diseases.

Q fever scientific opinion 2010 (<http://www.efsa.europa.eu/en/scdocs/doc/1595.pdf>)

Development of harmonised schemes for the monitoring and reporting of Q-fever in animals in the European Union (<http://www.efsa.europa.eu/en/scdocs/scdoc/48e.htm>)

Bee Mortality and Bee Surveillance in Europe (<http://www.efsa.europa.eu/en/scdocs/scdoc/27e.htm>)

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Article 36 project team Development of harmonised schemes for the monitoring and reporting of Q-fever in animals in the European Union: Sidi-Boumedine K, Rousset E, Henning K, Ziller M, Niemczuck K, Roest HJ, Thiéry R

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# Rotavirus in Human and Domestic Animals in Sudan

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## INTRODUCTION

In humans, rotavirus has been documented to cause about 111 million cases of gastroenteritis requiring home care annually, 25 million clinic visits, 2 million hospitalizations and a median of 440,000 deaths in children under 5 years of age world wide (Parashar *et al.*, 2003). Rotavirus was the major detected pathogen associated with diarrhea in Melit District in South Sudan (Sixl *et al.*, 1987).

In Sudan, Mohamed *et al.* (1998) detected rotavirus in 8 out of 200 camel calves; and by us in 13.9% of 332 camel calves with diarrhea (Ali *et al.*, 2005).

In Sudan, rotavirus was detected in 77 out of 116 calf samples (El Nour, 1994).

Berrios and colleagues (1988a) described the association of rotavirus infection with diarrhea in young goats.



## MATERIALS and METHODS

**Sample collection:** A total of 198 stool samples were collected during 2005-2006 from 85 infants, 50 camel calves, 40 cattle calves, and 23 goat kids.

**Group A rotavirus antigen detection:** All fecal samples were examined by Rotavirus IDEIA ( DAKO Diagnostics, U. K.).

**Polyacrylamide gel electrophoresis (PAGE):** 20 samples, were tested by PAGE according to published methods (Steele and Alexander, 1987).

## RESULTS

### Rotavirus antigen detection:

A total of 33 (12 infants, 13 cattle, 5 goat and 3 camel) samples tested for group A rotavirus were found positive (Fig. 1).

### Polyacrylamide gel electrophoresis (PAGE):

Of 20 samples examined by PAGE, 10 were positive for group A rotavirus Electropherotype with different profiles (Fig.1)

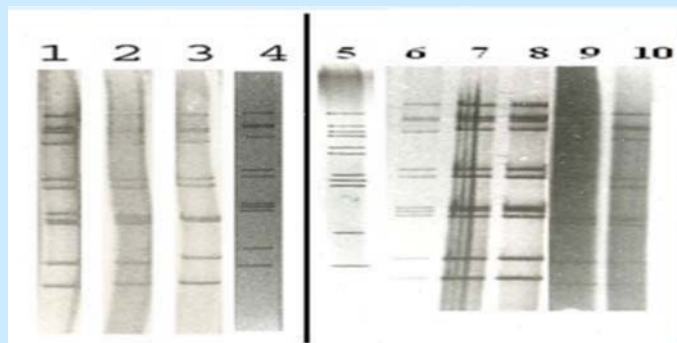


Figure 1 : Detection of group A rotavirus in fecal samples of infants and domestic animals in Sudan using ELISA.

Figure 2: Characteristic group A rotavirus long profile detected by PAGE, lane 1.2.3.4.10 human, lane 5 camel, lane 6,7,8,9 cattle samples

## DISCUSSION and CONCLUSION

In this study rotavirus antigen was detected in 14.1 % of 85 tested infants. In the present study, rotavirus antigen was detected in 6 % of 50 tested camel calves: this confirms the association of rotavirus with camel calf diarrhea.

The results obtained in this small study reveal the detection of rotavirus antigen in 32.5 % of 40 tested calves, this was the highest incidence noticed in this work and is higher than most of the previous reports. This study has shown the significant role of rotavirus in diarrhea in kids (21.7 %).

Characteristic group A rotavirus long profile was seen in 10 samples, most of them were human samples.

Group A rotavirus is existing in human and domesticated animals in Sudan.

The highest rotavirus incidence was in cattle then in goats.

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# The effect of neutering on the risk of mammary masses: a systematic review

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## Aim

The primary aims of this project were:

- > To evaluate the **strength of evidence** for an association between neutering, or age of neutering, and mammary masses of any histological type in bitches
- > To estimate the **magnitude of the association**.



## Method

A **systematic review** was conducted according to a pre-defined **protocol based on Cochrane guidelines**. PubMed<sup>a</sup>, Cab Direct<sup>b</sup> and ISI Web of Knowledge<sup>c</sup> were searched according to terms shown in Box 1. Articles from peer-reviewed journals published in English were eligible, as shown in Box 2.

Eligible articles were reviewed independently by two epidemiologists, using a pre-defined data extraction form based on suggestions from the Cochrane Handbook<sup>d</sup>. Risk of bias was assessed using the Cochrane<sup>d</sup> (trials), Newcastle Ottawa<sup>a</sup> (case control; cohort) and Downs and Black<sup>f</sup> (cross-sectional) systems and classified according to SIGN<sup>g</sup> recommendations.

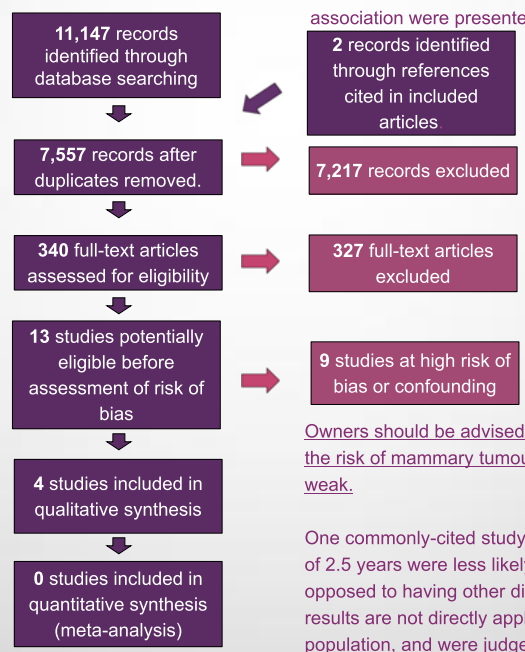
### Box 1. Search Terms

- Dog OR dogs OR bitch\* OR canis\* OR canine\* OR canid\* OR "Dogs"[MAJR]<sup>h</sup>
- Spay\* OR neuter\* OR ovariectomy\* OR ovarioectomy\* OR gonadect\* OR gonad OR gonads OR spey\* OR "Ovariectomy/veterinary"[MAJR]
- mammar\* OR breast\* OR "Mammary Glands, Animal"[MAJR]
- tumour\* OR tumor\* OR cancer\* OR neoplas\* OR mass OR masses OR lump\* OR "Neoplasms/veterinary"[MAJR]

(#1 AND #2) OR (#1 AND #3 AND #4)

### Box 2. Eligibility Criteria

- Either** the frequency of mammary masses was measured in both neutered and entire female dogs (or animals neutered at different ages); **or** the frequency of neutering (or neutering at different ages) was recorded in both female dogs with and without a history of mammary masses.
- The results of Criterion 1 were reported.
- The "neutered" dogs were neutered by ovariectomy or ovariohysterectomy.
- The report was an original research article.<sup>i</sup>
- The report was published in a peer-reviewed journal (according to the journal's current guidelines).<sup>i</sup>
- The full text of the report was available in English.<sup>i</sup>



## Results

Of 11,149 records initially identified by the search strategy, 13 articles satisfied the eligibility criteria, as shown in the flow-diagram. Nine of these were judged to have a high risk of bias; the remaining four<sup>1,2,3,4</sup> were classified as having a moderate risk of bias. The table below summarises potential bias in included studies.



**One study<sup>1</sup> of histopathology results (n=174 found a strong protective effect of neutering before 2.5 years of age (Relative Risk 0.06), particularly neutering before 1<sup>st</sup> oestrous (Relative Risk 0.005) on the risk of mammary malignancy, as opposed to other diagnoses. However, these results are not directly applicable to the general dog population because the study population consisted only of dogs from whom biopsies were submitted. Also, no confidence interval or p value was presented and there were other potential biases, summarized in the table below.**

**Two studies found no evidence of an association between neutering and the risk of developing any mammary mass<sup>2</sup> (n=144) or the risk of neoplasia amongst dogs with mammary masses<sup>3</sup> (n=2270). One study<sup>4</sup> (n=65) reported "some protective effect" of neutering on the risk of developing mammary tumours, but no measures of association were presented.**

		Bias in Included Studies						
Source of Bias <sup>j</sup>		Confounding	Control Selection	Missing Data	Neuter Status Ascertainment	Case Selection	Miscellaneous	Loss to Follow Up
Case Control	Schneider 1969	High Risk	High Risk	High Risk	High Risk	High Risk	High Risk	High Risk
	Perez Alenza 1998	High Risk	Unclear Risk	High Risk	High Risk	High Risk	High Risk	High Risk
	Richards 2001	High Risk	High Risk	High Risk	High Risk	High Risk	High Risk	High Risk
Cohort	Bruenger 1995	High Risk	High Risk	High Risk	High Risk	High Risk	High Risk	High Risk

**Legend**

High Risk of Bias	High Risk of Bias
Low Risk of Bias	Low Risk of Bias
Unclear Risk of Bias	Unclear Risk of Bias

j. The studies were at low risk of bias in the following categories:- case control: case definition, miscellaneous; cohort: length of follow up, classification of mammary tumour status and ensuring cohorts were disease-free at start of study. k. Two different control groups used.

## Conclusions

Owners should be advised that the overall strength of evidence that neutering reduces the risk of mammary tumours—and that early neutering is more strongly protective—is weak.

One commonly-cited study of biopsy results found that bitches neutered before the age of 2.5 years were less likely to be diagnosed with malignant mammary tumours, as opposed to having other diagnoses (reduction approximately 20-fold). However the results are not directly applicable to the risk of developing tumours in the general population, and were judged to be at moderate risk of bias and confounding. Two studies, also at moderate risk of bias, found no evidence that neutering either reduces the risk that a dog will have a mammary mass, or reduces the risk of malignancy amongst dogs with mammary masses.

This information should be balanced with other available information on the risks and benefits of neutering in general, and in particular early versus late neutering.

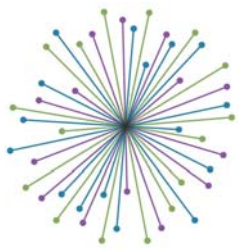
Further research into the association between mammary masses and neutering should focus on recording:

- > age, breed and exposure to synthetic Progesterones, as potential confounders.
- > age at neutering and length of subsequent follow-up period.

## Acknowledgements

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# Caseload of final year veterinary students: Collecting evidence



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CENTRE FOR EVIDENCE-BASED  
VETERINARY MEDICINE  
Putting research into practice

- ❖ Final year veterinary students at The University of Nottingham undertake a series of rotations through various veterinary disciplines and species-specific practices prior to graduating and entering the profession
- ❖ In a community-based education model where students undertake rotations through Clinical Associate practices, it is difficult to assess the caseload seen by students during rotations. It is important to ensure they have had the opportunity to participate in many facets of veterinary practice during their final year of training
- ❖ There is very little published information outlining the types of cases commonly presented to veterinary surgeons in the UK, which makes determining whether final year students are seeing the appropriate caseload difficult
- ❖ Documentation of the cases seen by students is a way of assessing rotation content, and additionally adds to the evidence-base in veterinary medicine on the common conditions seen in practice

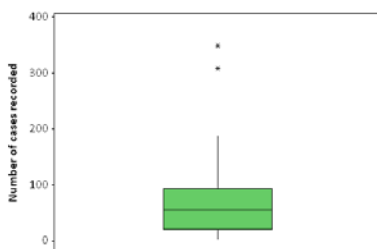
## Methods:

Final year veterinary students at the School of Veterinary Medicine and Science at The University of Nottingham were asked to voluntarily record information on cases they were involved in during rotations.

This was done using a basic clinical coding booklet, and an electronic database located within an online site already used by students for general communication, feedback and assessment whilst on rotations.

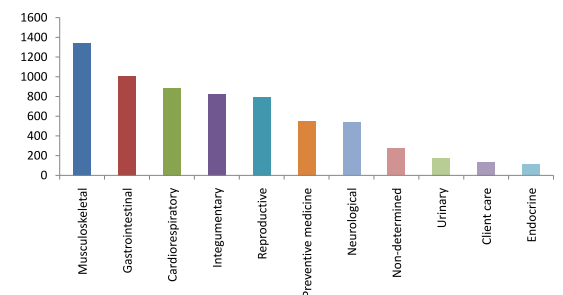
Seventy eight of 87 final year students recorded case information electronically, with a total of 5432 cases recorded between May 2010 and February 2011.

The data were not normally distributed, with a median of 57 cases recorded per student (IQR 23-93).

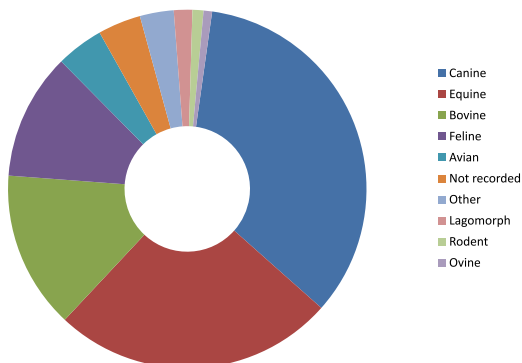


Of the cases recorded detailing diagnoses (n=2568/5432; 48%), most did not state that a definitive diagnosis had been reached (n=1892; 74%).

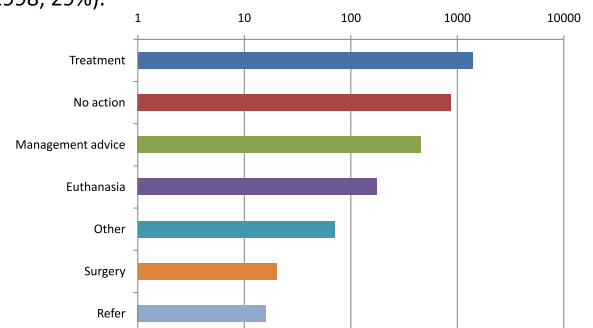
Of the total number of entries for body system (n=6642), the majority of recorded cases involved the musculoskeletal (20%), gastrointestinal (15%) and cardiorespiratory (13%) systems.



Of the total entries for species (n=5432), the majority of cases involved canines (34%), followed by equines (25%) and bovines (14%).



Of the recorded cases selecting one of the outcome options (n=2998/5432; 55%), the majority of cases resulted in some form of treatment (n=1388/2998; 46%), followed most commonly by no treatment (n=870/2998; 29%).



## Conclusion:

These results show that a broad range of cases involving a variety of body systems and species were recorded by most final year veterinary students during their rotations.

The information here adds to the limited knowledge base on what vets are commonly presented with in veterinary practice and how these cases are managed.

More detailed information is being collected as part of the research conducted by the Centre for Evidence-based Veterinary Medicine on cases seen in veterinary practice and conditions seen commonly by veterinary surgeons in the UK.

## Acknowledgements:

Many thanks to the students and staff at SVMS for making this study possible and to The University of Nottingham and Novartis Animal Health for sponsoring this research.

# A survey of current farming practices in England and Wales relating to the use of antimicrobials in dairy cattle and feeding of waste milk



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## Introduction

In recent years bacteria, particularly *E. coli*, have emerged with resistance to 3<sup>rd</sup> and 4<sup>th</sup> generation cephalosporin antibiotics. These antibiotics are widely used as front line treatments in human medicine and the development and dissemination of bacteria resistant to these medicines is a serious medical problem. This resistance is mainly conferred by the production of enzymes such as Extended Spectrum Beta Lactamases (ESBLs). Ongoing surveillance studies at the Veterinary Laboratories Agency have observed farms where ESBL *E. coli* are present and detected more frequently in young calves than older cattle. Calves are sometimes fed waste milk, which is milk that cannot be sold for human consumption. Waste milk might include colostrum, mastitic milk and milk from cows treated with antibiotic or non-antibiotic medicines that may lead to a residue in the milk. The purpose of this survey was to assess current practices for the antibiotic treatment of dairy cows and feeding of waste milk to dairy-bred calves on dairy farms in England and Wales.

## Methods & Response Rate

A sampling frame of dairy farms in England and Wales stratified by herd size was constructed from the Cattle Tracing System database and 4000 farms randomly selected from this. Each farm was sent a postal questionnaire and covering letter explaining the purpose and background to the study. A sample size calculation showed us that at least 385 responses were required to allow us to estimate with 95% confidence what proportion of farms were carrying out these practices.

In total 569 farms responded to the survey, of which 527 (93%) kept dairy cows and reared calves to weaning and so could complete the questionnaire in full (Fig. 1). A summary of the key findings from the survey are presented below.

## General Farm Characteristics

Of the 527 farms that completed the questionnaire, 81% did feed waste milk to calves. We had responses from 48 different counties across England and Wales with the highest proportion of results from Devon (11%) and Cumbria (8%) - counties known to have some of the highest cattle populations in Great Britain. The herd size of respondents (adult milking cows) ranged from 1 to 1000 with a mean herd size of 154 (Fig. 2).

The majority of respondents (74%) calved all year round. Of those that calved seasonally, autumn was the most common season for calving (13%) followed by spring (8%).

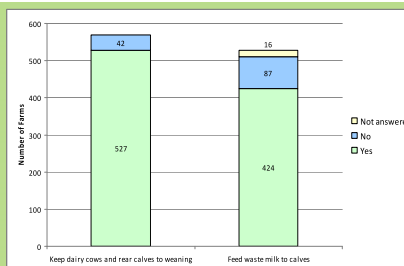


Figure 1 A summary of responses to the survey

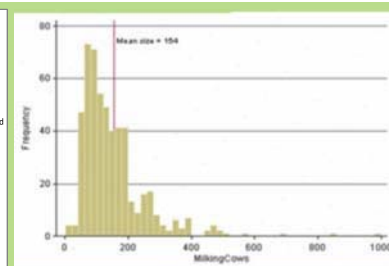


Figure 2 Distribution of the sizes of milking cow herds on farms which took part in the survey

## Antibiotic Treatments

The number of cases of mastitis treated during lactation in the last year ranged from 0 to 500 with a mean of 47 (Fig. 3). Around 90% of respondents used antibiotic tubes to treat mastitis. Cefquinome, a 4<sup>th</sup> generation cephalosporin, was the second most frequently used (Fig. 4). 96% of respondents used dry cow antibiotic tubes at drying off. Cefalonium, a 1<sup>st</sup> generation cephalosporin, was most frequently used by 43% of farms (Fig. 5).

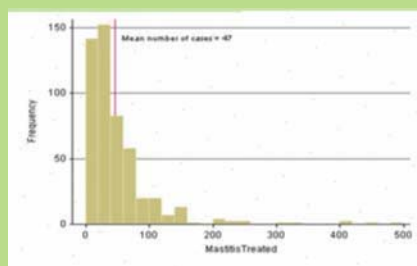


Figure 3 Distribution of the number of mastitis cases in dairy herds

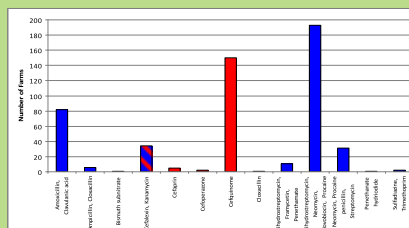


Figure 4 Frequency of milking cow tubes used to treat mastitis in lactating cows, grouped by active ingredients. Red bars indicate cephalosporins (patterned bar = cephalosporin in combination with other antibiotics)

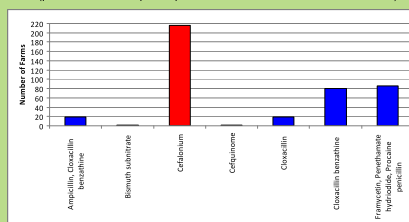


Figure 5 Frequency of dry cow tubes used at drying off, grouped by active ingredients. Red bars indicate cephalosporins.

The most commonly used injectable antibiotics were Tylosin (27% of farms) Dihydrostreptomycin & Procaine penicillin (20% of farms) and Ceftiofur, a 3<sup>rd</sup> generation cephalosporin (13% of farms) (Fig. 6). On average, respondents used their most commonly used injectable antibiotic in 18% of their herd.

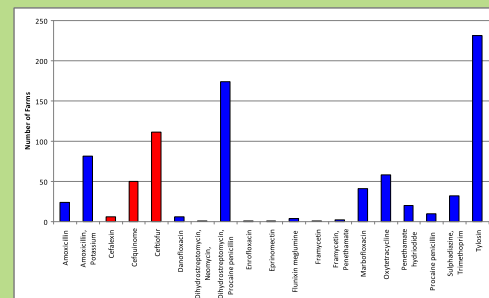


Figure 6 Frequency of injectable antibiotics used in dairy cows. Red bars indicate cephalosporins.

## Waste Milk

This survey revealed that the majority of farms leave calves to suckle for up to 24 hours after birth. The least common practice was to remove calves straight away although this did occur on around 15% of farms.

As shown in Fig. 1, 81% of respondents did feed waste milk to calves. The survey also revealed that 356/410 respondents fed milk to calves from cows with mastitis (87%). Fig. 7 illustrates that the majority of waste milk contained milk or colostrum from cows in receipt of antibiotics.

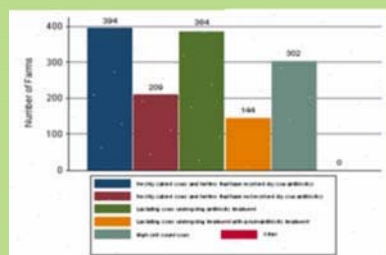


Figure 7 Description of the contents of waste milk on farms which took part in the survey. Multiple answers could be given so individual farms may be represented more than once.

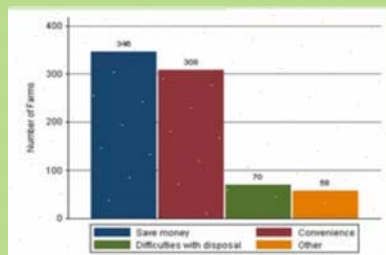


Figure 8 Reasons given by farms for feeding waste milk. Multiple answers could be given so individual farms may be represented more than once.

The most common factor affecting respondent's decision to feed waste milk to calves was to save money, closely followed by convenience (Fig. 8). Other factors that were specified generally consisted of improvements in calf growth and immunity.

## Further Work

This survey has provided a great deal of data on current farm practices in England and Wales which has not been available before. The key data obtained has been presented here and shows that the feeding of waste milk from cows in receipt of antibiotics is common in England and Wales. Further work is underway to assess the levels of antibiotic residues in waste milk, and to determine whether these can select for ESBL *E. coli*. That information, in combination with the data obtained by this survey will be used to develop a risk model to assess the risk of the acquisition or selection of ESBL *E. coli* from feeding calves waste milk and to recommend practical control measures.



# Trade contact network in the pork supply chain

## Characterisation of the network topology

### Data basis

- ◆ 176 premises from a producer community in Northern Germany
- ◆ 6,892 animal movements in an observation period from 2006 – 2009  
→ 770 different trade connections

### Static network analysis

#### In-degree

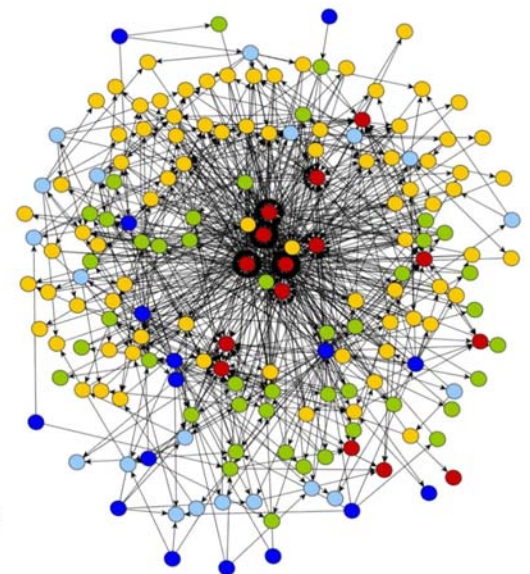
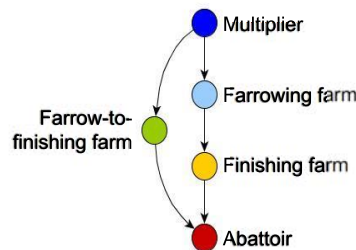
- ◆ Number of trade partners which **deliver** animals to a specific premise

#### Out-degree

- ◆ Number of trade partners which **receive** animals from a specific premise

#### Scale-free networks

- ◆ Power-law degree distribution →  $p_k \sim Ck^{-\alpha}$
- ◆ High amount of low-degree premises, small number of high-degree premises

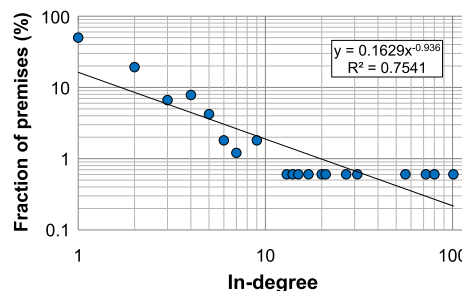


### Results

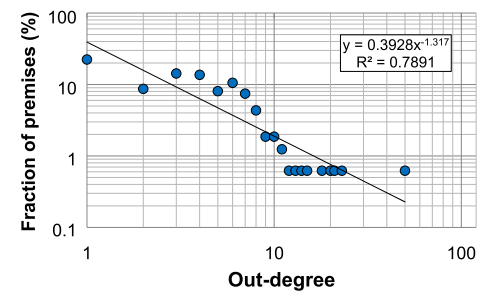
#### In- and out-degree per premise type

Premise type	n	In-degree			Out-degree		
		25 <sup>th</sup> Pctl	50 <sup>th</sup> Pctl	75 <sup>th</sup> Pctl	25 <sup>th</sup> Pctl	50 <sup>th</sup> Pctl	75 <sup>th</sup> Pctl
Multiplier	16	0	0	1	3	5	10
Farrowing farm	20	1	2	3	1	3	8
Finishing farm	81	1	2	3	2	3	5
Farrow-to-finishing farm	44	1	1	2	2	5	7
Abattoir	15	6	20	56	0	0	0
<b>Total</b>	<b>176</b>	<b>1</b>	<b>1</b>	<b>3</b>	<b>1</b>	<b>4</b>	<b>6</b>

#### In-degree distribution

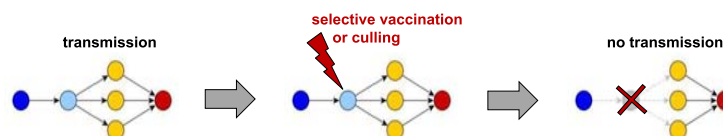


#### Out-degree distribution



### Conclusion

- ◆ Network analysis provides a substantial tool for characterising contact patterns
- ◆ Different types of premises reveal various degrees reflecting their position in the pork supply chain
- ◆ The described network shows scale-free characteristics
- ◆ Scale-free networks are highly resistant concerning the random removal of premises



- By strategic removal of highest-degree premises the network structure changes and can decompose into fragments.
- The chain of infections can be interrupted and further disease spread can be prevented.

# Unexpected Delay in Transmission between Spatially Separated Hosts

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N.M. Bolder<sup>1</sup>, G. Nodelijk<sup>1</sup>, M.C.M. de Jong<sup>2</sup>

## Introduction

Neighborhood infection is an important route for the outbreak of many (pathogenic) diseases. Lately it has received much attention because of epidemics of Avian Influenza and other pathogens, resulting in severe economical and animal losses. In this study we used the data from indirect transmission experiments to develop and test a new class of models for indirect transmission.

## Aim

Gain more insight into the underlying mechanisms of indirect transmission.

## Methods

Different scenarios from a newly developed class of models were tested against the data from indirect transmission experiments, using a maximum likelihood approach. Estimated parameter values need to be biologically plausible.

## Experimental setup



Indirect transmission of *Campylobacter* between broilers used as model system.

## Tested scenarios

Pure airborne



Pathogen heterogeneity



One environmental compartment



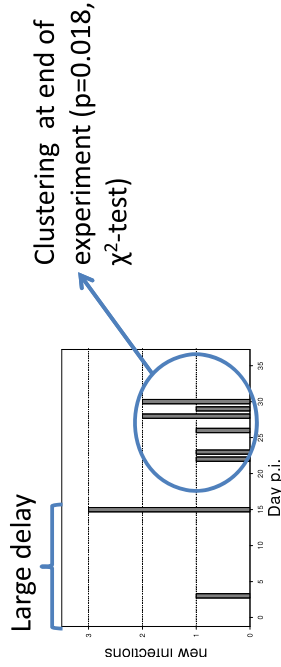
Two staged environmental compartments



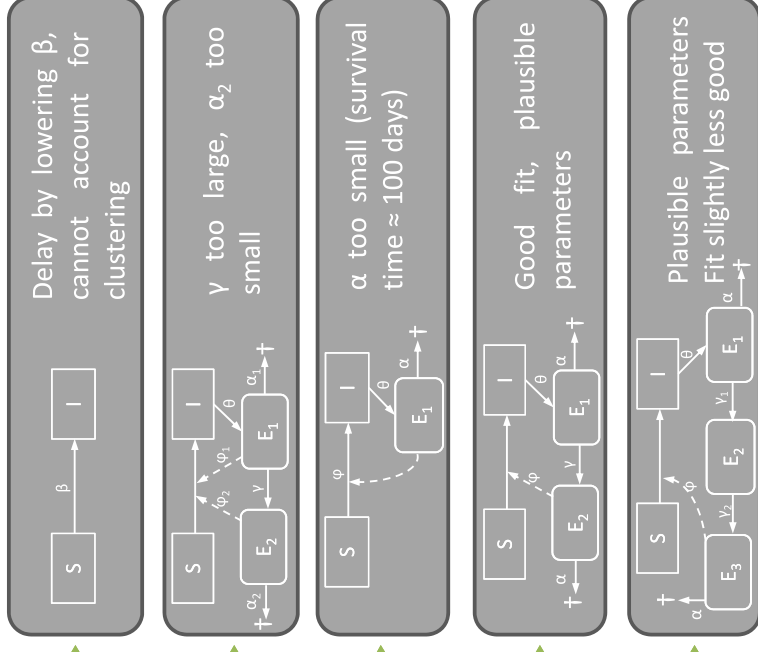
Three staged environmental compartments



## Data



## Results



## Conclusion:

- Indirect transmission cannot be explained by 'pure' airborne transmission
- Multistep staged route from source to recipient explains the data best.

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# A case-control study of risk factors for bovine cysticercosis in Danish cattle herds



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## Objective

To quantify associations between potential risk factors and bovine cysticercosis in dairy and beef cattle at herd level for potential use to select animals from high-risk herds for future risk-based meat inspection in Denmark

## Materials and Methods

- 308 herds included in the study
  - 202 dairy and veal producers. 49 case herds, 153 control herds
  - 106 beef producers. 28 case herds, 78 control herds
- Case herd definition
  - herds hosting at least one animal diagnosed with bovine cysticercosis at meat inspection for >100 days in period January 2006 to March 2010 (n=77)
- Control herd definition
  - herds not hosting any animals diagnosed with bovine cysticercosis at meat inspection in the period January 2004 to March 2010
  - three control herds were selected per case herd by random sampling, stratified on dairy and beef herds (n=231)
- Farm managers interviewed by telephone regarding cattle production, management practices, farm location, staff and visitors
- Univariable analyses and multivariable logistic regression analysis performed with forward inclusion of variables  
 Significant interactions only kept in model if meaningful. Significance level was set to 5% in regression analysis



Fig. 1 Distribution of bovine cysticercosis case herds, potential case herds, selected control herds and other active cattle herds in Denmark from 2006 to 2010. No obvious clustering.

## Results

Animals grazing and farming type  $P = 0.037$



Levels	OR	95% CI
All animals grazing, organic	2.5	1.1 - 6.0
All animals grazing, conventional	2.0	1.0 - 4.1
Non or some animals grazing, conventional	1.0	

Herd size  $P < 0.0001$



OR=1.5, 95% CI: 1.2 - 1.8  
 per 100 animals increase in herd size

Persons with daily access to farm area >50 yrs old  $P = 0.013$



OR=2.4, 95% CI: 1.2 - 4.8  
 compared to herds where persons with daily access were below 50 years old

Higher risk of having infected people on farm?  
 Or behavioural difference?.

## RISK FACTORS FROM MULTIVARIABLE MODEL



Access to risky water source (stream, river or surface water) and  
 Sewage treatment plant in proximity  $P = 0.002$

OR= 2.5, 95% CI: 1.1 - 5.6  
 compared to herds in which there are no sewage treatment plants in the proximity



Share machinery or hire contractors  $P = 0.028$



OR= 5.7, 95% CI: 0.9 - 36.5  
 compared to herds not using shared machinery  
 Note that very few herds did not use shared machinery  
 Not feasible factor in a risk-based surveillance system

## Conclusions and Perspectives

- This study supports previous studies where cattle drinking from risky water source (usually surface water) in combination with having a sewage treatment plant in the proximity of the farm were major risk factors
- Grazing was significantly associated with finding carcasses with bovine cysticercosis at meat-inspection, regardless of farming type indicating that using organic farming for selection of high-risk herds is most likely insufficient  
 Food chain information might be useful to record cattle that have been grazing.
- There is no clear reason why persons >50 yrs old are more risky on farm, but it might be related to higher risk of being infected with *Taenia saginata* or it is an indicator of certain unknown farming practices
- Large herds are more likely to have at least one test-positive animal identified at meat inspection probably due to increased herd level sensitivity when testing more animals. Analysis of animal level data might add more information for risk-based systems.



# The relationship between herd size and milk somatic cell counts of Irish dairy cows: A retrospective cohort study

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## Introduction

Within the EU25, Ireland was the 8<sup>th</sup> largest producer of cows milk, producing 5.2 million tonnes of milk. Ireland is one of the worlds leading producers of infant nutrition products, producing 15% of the worlds powdered infant formula (IBEC, 2007), which requires low somatic cell count (SCC) milk.

Somatic cell counts are the single most important indicator of milk quality, elevated milk SCC above 200,000 cells/ml are usually indicative of infection.

Within the EU, commission regulations state that food operators must initiate procedures if raw bulk tank milk SCC exceed 400,000 cells/ml (EC, 2006; rolling geometric mean over 3 months).

## Objectives

The objectives of this study were to determine the relationship between herd size and;

- Geometric mean individual cow milk SCC
  - above 200,000 cells/ml (*MSSC200*) over an entire lactation
  - above 400,000 cells/ml (*MSSC400*) over an entire lactation.
- First test day milk SCC (cows ≤ 50 days in milk)
  - above 200,000 cells/ml (*50SCC200*)
  - above 400,000 cells/ml (*50SCC400*)

## Materials and Methods

Individual cow monthly milk records for all cows in Ireland that were milk recorded in 2009 were obtained from the Irish Cattle Breeding Federation (ICBF) and data on herd size was obtained from the Department of Agriculture, Fisheries and Food, Animal Identification and Movement system (AIM).

Data was restricted to cows with ≥7 monthly records in order to calculate geometric mean SCC.

Data was managed using Microsoft Excel<sup>®</sup> and analysed using STATA<sup>®</sup>. Maps were generated using ArcGIS<sup>®</sup>.

Descriptive analyses were performed and individual herds were mapped to visually identify their location relative to the national dairy cattle population density (Figure 1).

Data were analysed using multivariable logistic regression models, with random effects to account for clustering of cows within herds.

## Results

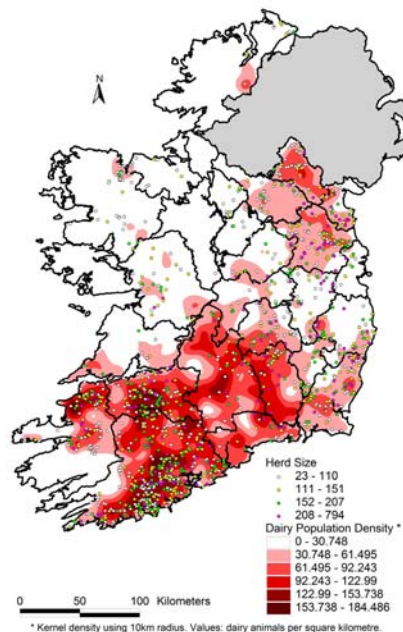


Figure 1: Locations of herds used in this study relative to the dairy cattle population density in Ireland.

- 26% and 11% of cows had *MSSC200* and *MSSC400*, respectively.
- 22% and 13% of cows had *50SCC200* and *50SCC400*, respectively.
- *MSSC200* was associated with cows from larger herds
  - 152 to ≤ 207 V's ≤ 110 cows; OR 1.24, 95% CI 1.09-1.14, P=0.001
  - ≥ 208 cows V's ≤ 110 cows; OR 1.36, 95% CI 1.18-1.58, P<0.001
- *MSSC400* was associated with cows from larger herds
  - 152 to ≤ 207 V's ≤ 110 cows; OR 1.29, 95% CI 1.12-1.48, P<0.001
  - ≥ 208 cows V's ≤ 110 cows; OR 1.42, 95% CI 1.21-1.66, P<0.001
- *50SCC200* was associated with cows from larger herds
  - 148 to ≤ 200 V's ≤ 107 cows; OR 1.15, 95% CI 1.04-1.28, P=0.01
  - ≥ 201 cows V's ≤ 107 cows; OR 1.30, 95% CI: 1.16-1.46, P<0.001
- *50SCC400* was associated with cows from larger herds
  - 148 to ≤ 200 V's ≤ 107 cows; OR 1.20, 95% CI 1.07-1.34, P<0.001
  - ≥ 201 cows V's ≤ 107 cows; OR 1.34, 95% CI 1.18-1.52, P<0.001

## Conclusions and future work

There is an association between herd size and milk SCC, with implications for both animal health and the marketability of Irish milk.

These results are important given Ireland's desire to increase milk supplies by 50% by the year 2020 (DAFF, 2010).

Further work is needed to examine the impact of herd expansion, not just herd size, on SCC.

# ASSESSMENT OF BIOAEROSOLS IN SWINE BARN IN PORTUGAL



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## BACKGROUND & OBJECTIVE

Poor air quality in livestock buildings adversely affects animal health through a variety of mechanisms, and also enables the transmission of diseases, like Salmonellosis. In bioaerosols we have bacteria and fungi. The Gram negative bacteria (G-) are some of the most important pathogens for swine, and included fecal bacteria, like *Escherichia coli*, *Salmonella* sp., *Neisseria* sp., *Pseudomonas* sp., among others. Some studies have reported G- bacterial exposures in swine barns between  $7 \times 10^3$  and  $65 \times 10^5$  CFU/m<sup>3</sup> (1), while others (2) reported significantly lower concentrations of  $0.42$  to  $4.52 \times 10^2$  CFU/m<sup>3</sup>. Salmonella airborne transmission was proved possible by experimental studies (3,5). The infection dose found were higher than  $10^6$  CFU and depends of serotype.

**AIM:** To assess the bioaerosol contamination in swine barns and with this to obtain field information providing an insight into the possible risk of airborne transmission of Salmonella in swine herds. For this purpose **Gram negative bacteria (G-)** were used as an **indicator** of possible Salmonella presence.

## MATERIALS & METHODS

### DATA:

- ❖ One herd with post-weaning barns (active ventilation) and two types of fattening barns (passive versus active ventilation).
- ❖ Samples taken at the passages in the middle of the barn at a height near the nostril of the pigs.
- ❖ An air volume of 5 liters was aspirated to total bacteria counts and fungi and 30liters to G- counts.
- ❖ Samples taken once a month in January, February and April 2009 (winter to spring days).
- ❖ Temperature and humidity at the barn and outside the barn was recorded using a hygrometer.
- ❖ Potential differences between barns and months were tested.

### MICROBIOLOGICAL ANALYSIS:

- ❖ The air was aspirated by “air IDEAL microbiological air sampler” (bioMérieux®) to 90mm diameter Petri dishes.
- ❖ Three types of culture media were used: Agar MacConkey for G-, Agar Gelose Tripcase Soja for total bacterial and Agar Sabouraud glucose cloranfenicol for fungi.
- ❖ The plates were incubated at 30°C/72h to total bacteria, at 37°C/48h to G- and at 25°C/5days (120h) for fungi.
- ❖ CFUs counts were corrected to most probable number of bacteria (MPN) and divided by the volume of air sampled and are presented as CFU/m<sup>3</sup>.

## RESULTS & DISCUSSION

- Mean stocking densities were 20 pigs/pen in fattening pens (pens with 4x4m) and 25 pigs/pen in post-weaning pens (pens with 2x2m).
- Figure 1 and 2 shows the distribution of CFU/m<sup>3</sup> fungi, total bacteria and G- for naturally and mechanically ventilated fattening barn and post-weaning barn.
- There were **no statistical difference** between barns for fungi, total bacteria and G- counts .
- Comparing the values of CFU/m<sup>3</sup> of fungi along the months tested showed statistical differences ( $p < 0.04$ ) between January and the rest of the months (Figure 3).

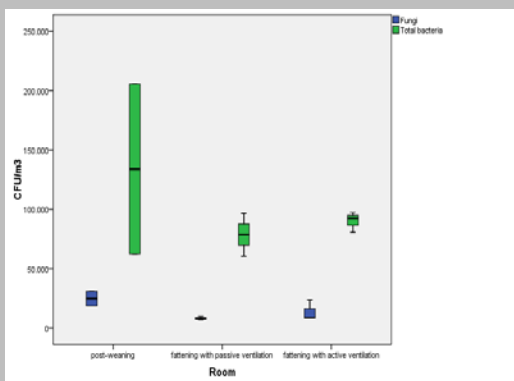


Figure 1: Distribution of CFU/m<sup>3</sup> of total bacteria and fungi between barns

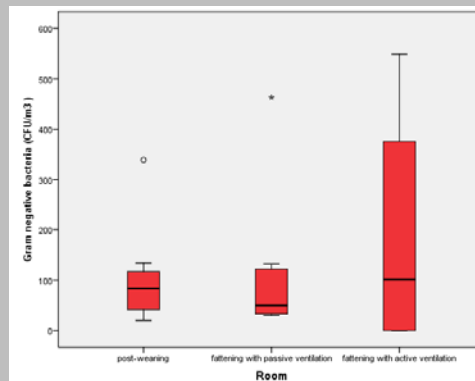


Figure 1: Distribution of CFU/m<sup>3</sup> of G- bacteria between barns

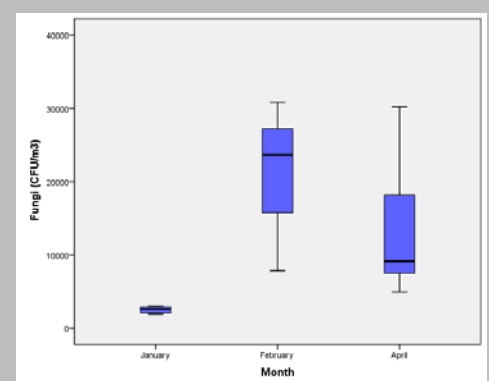


Figure 1: Distribution of CFU/m<sup>3</sup> of fungi between months

## CONCLUSIONS

- ✓ **High levels of total bacteria and fungi** concentration in all barns, similar to the ones found in others studies (4).
- ✓ This indicate **a highly contaminated air** with could favor the transmission of microbiological agents within the farms. The piglets are especially at risk because they have a poor immunity system and the results in the post-weaning barn were worst than in the others barns.
- ✓ The contamination with **G- was lower than expected**. It is lower than the one found in other studies (1) but agrees to with Chang's data (2).
- ✓ Using the G- bacteria counts as indicator of fecal contamination and of the possible presence of *Salmonella*, the concentration of *Salmonella* (if present) will be in lower than G- counts and under the infective dose of *Salmonella* trough the air, therefore the risk of airborne infection with *Salmonella* seemed negligible.
- ✓ Although the number of samples from the study was low we decided not to continue with this study because we felt that the results would not become different then the ones obtained.

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### Acknowledgments:

SVEPM's contribution to the delegate conference attendance bioMérieux® Farmer and veterinarian that participated in this study

# MODELLING TRANSMISSION PARAMETERS FOR *SALMONELLA* TYPHIMURIUM IN SWINE USING A BAYESIAN APPROACH



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<sup>2</sup> Segalab – Laboratório de Sanidade Animal e Segurança Alimentar, Rua de Recarei, Gondival, 4465-734 Leça do Balio, PORTUGAL

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## BACKGROUND & OBJECTIVE

*Salmonella* Typhimurium is a current major food-borne pathogen in humans and mainly associated with pork and pork products. At herd level testing is a part of the control programs for this agent. Modeling the transmission of this agent in the herd is one way of understanding the factors that influence the dynamic of transmission and may contribute to improve the efficiency of *S. Typhimurium* control programs.

**AIM:** to estimate transmission parameters ( $\alpha$ ,  $\delta$  and  $\beta$ ) for *S. Typhimurium* in swine herds using field data from a cohort study of infection using a three state (SIR) transition model.

## MODELLING APPROACH

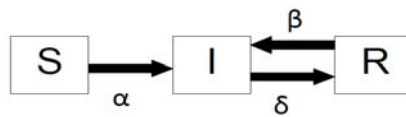
### DATA:

- 6 cohorts of 30 pigs (2 cohort per herd) followed since 4 weeks old until the slaughter age.
- The herds were known to be infected with *S. Typhimurium*.
- The pigs were tested with intervals of 3 to 4 weeks for presence of *Salmonella* in feces and sera.
- Pigs were classified as infected (I), susceptible (S) or resistant (R) according to the results of the tests (interpretation as parallel testing).

### ASSUMPTIONS:

- An infected pig would shed at least 4 weeks;
- Intermittent shedding (R→I);
- No transition of R to S;
- Markovian transitions;
- Specificity for both tests of 100%;
- All cohorts have infected animals in the beginning.;
- Time interval of 2 weeks.

### EPIDEMIC MODEL:



S – susceptible  
I – infected shedding  
R – infected not shedding

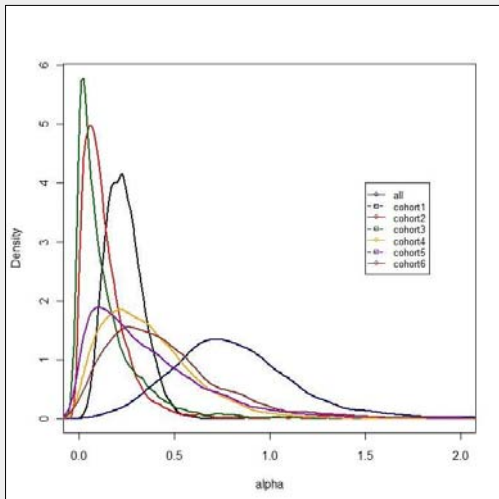
### BAYESIAN FRAMEWORK:

Poisson distribution for states.  
Modelling temporal dynamic of observed status of pigs in cohorts.  
Real health state of I depends of parallel Sensitivity of the tests.  
Inference on transition parameters between states.

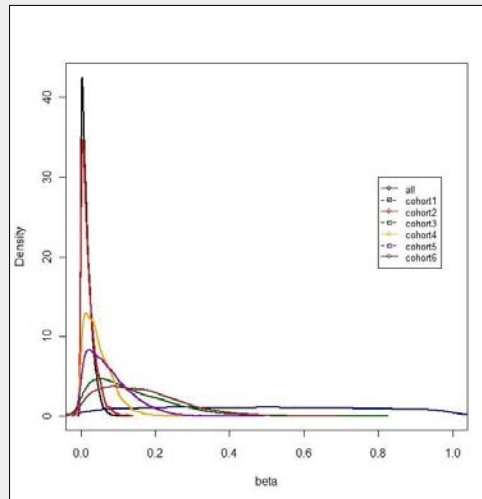
### PRIOR DISTRIBUTIONS:

Minimally informative priors for transmission parameters.  
Informative priors for sensitivity of both tests.

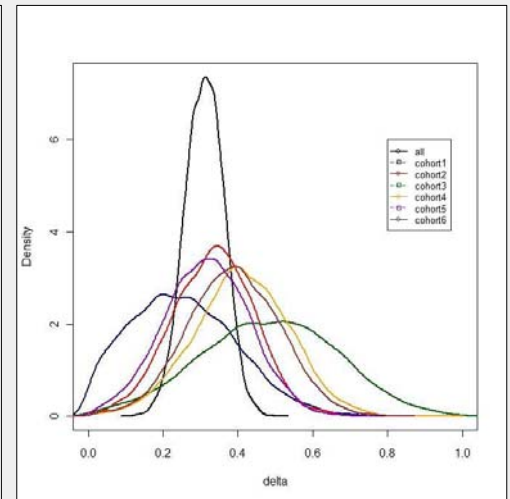
## RESULTS & DISCUSSION



Alpha: Transition rate from S to I for all data and for each cohort



Beta: Transition rate from R to I for all data and for each cohort



Delta: Transition rate from I to R for all data and for each cohort

- Transmission rate parameter alpha differs between cohorts, cohort 1 reach the highest values. Two groups (cohort 2 and 3) and (cohort 4,5,6 and 1) seem to be present.
- Transition between R to I (intermittent shedding) seems to be low and is quite similar for all cohorts except for cohort 1.
- Transition between I to R (stop shedding) is high and very similar between cohorts with the mode values around 0.3 for each 2 weeks.

## CONCLUSIONS

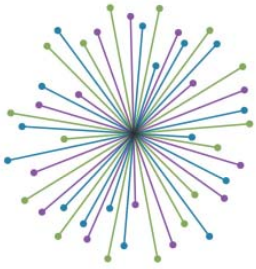
This study provides valuable information for future simulation studies such as:

- As the tests are not perfect for detection of *Salmonella* the sensitivity information should be incorporated in the models which use field data to assess better the real number of animals infected;
- As expected the dynamic of infection of this agent differs between cohorts of the same herds, so when simulating this aspect should be incorporated in the model;
- This information could be used for assessment of effectiveness of different control measures.

### Acknowledgments:

SVEPM contribution to the delegate conference attendance

Danish Agriculture and Food Council for the data



**CENTRE FOR EVIDENCE-BASED VETERINARY MEDICINE**  
Putting research into practice

# Practice-based research: what do vets want?

**School of Veterinary Medicine and Science**



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## Background

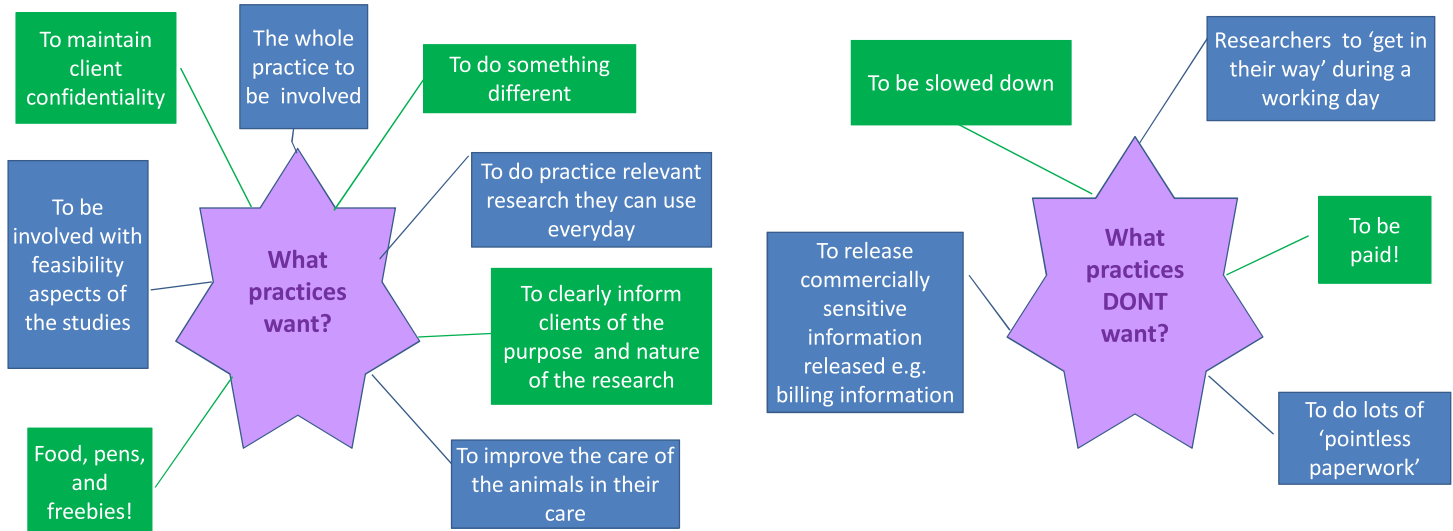
- Every day veterinary surgeons in first opinion small animal practice are presented with many animals that have naturally occurring disease
- Data is routinely collected, processed and archived by practices about their patients.
- This source of data is large and limitless, yet its value is only just starting to be realised. To date only small amounts of these data have been accessed for the purposes of veterinary research.
- A successful working relationship with first opinion veterinary practices is the key to:
  1. Undertaking research that is relevant to clinical practice
  2. Effectively accessing practice data
  3. Performing large scale observational studies and clinical trials
  4. Developing methods for delivering results of the research back to the consulting room
  5. 'Putting research into practice'

## Sentinel practices

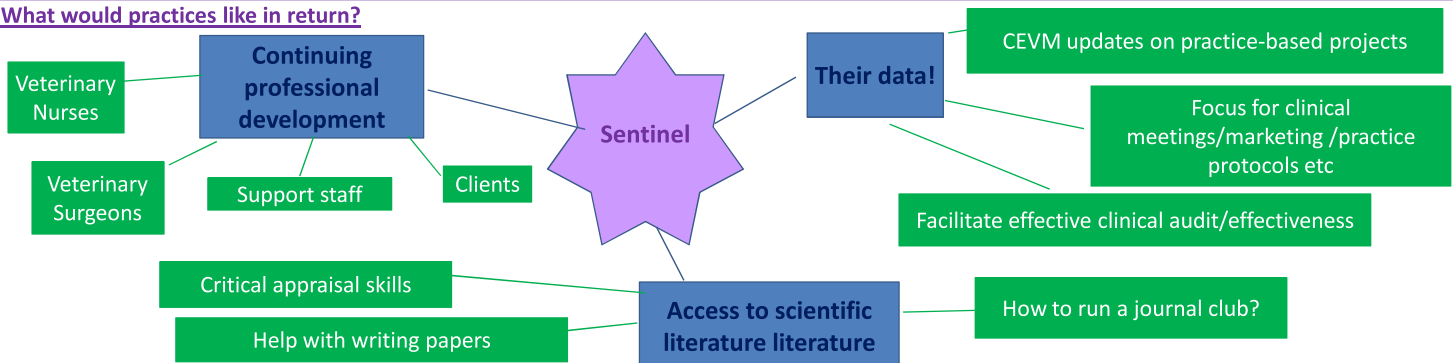
- A network of eight first opinion small animal practices has been established by the Centre for Evidence-based Veterinary Medicine (CEVM).
- These practices will be used to gather data for the first studies undertaken by the CEVM.
- During the development of the CEVM, these practices have been consulted regularly about various aspects of the research.
- Important factors that may help or hinder the research have been discussed, as well as what added benefits the practices could gain from being one of our sentinels.

What do vets want?  
How can you 'put research into practice' in a practical, feasible fashion?

## What practices have told us.....



## What would practices like in return?



## Conclusion

Continued effective communication between sentinel practices and researchers within the CEVM is the key to the success of this working partnership.

## Acknowledgements

Thank you very much to The University of Nottingham and Novartis Animal Health for sponsoring this research.

# Incorporating behavioural feedback effects into animal disease surveillance: a proposed framework

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## Introduction

Effective surveillance of endemic disease prevalence and monitoring for exotic disease incursion can be extremely resource intensive. The financial support to such systems is also often limited, so it is crucial that the resources available be allocated in the most efficient manner possible. Modelling of animal disease surveillance systems allows the effectiveness of different surveillance systems to be quantified, and therefore facilitates the optimisation of resource allocation. While heterogeneity between farms, for example in terms of animal numbers, is known to affect prevalence estimates and therefore is frequently incorporated into these models, it is much more difficult to incorporate behavioural effects which modulate the responses of individual farms to the perceived state of other farms around them. One solution is to utilize an agent-based approach, where the agents are further subdivided into actors representing farms of various types, and regulators such as media, government and veterinary interventions which have a feedback effect onto these actors. Each agent class is associated with a set of rules based on behavioural phenomena such as the perceived threat of disease incursion, and impacting on the probability that actors will look for disease. A major advantage of this approach is the ability to encapsulate different aspects of the system (for example an on-farm disease model, a between-farm movements model, and a behavioural feedback model) with only a few well defined interactions between actors and regulators. The internal functionality of each class can therefore be extensively re-written without the requirement to modify any other classes, or the code used to run the model.

## Model Framework

We present an object-oriented programming framework for an agent-based, stochastic model written in the C++ programming language. At the core of this model are objects representing a population of actors, and one or more objects representing regulators with feedback effects onto their behaviour. At each time step, actors pass information about their observed state to regulators (incorporating the sensitivity and specificity of the diagnostic tests), and receive information about the wider population in return. The generic actor and regulator classes derive to more specific classes, which add functionality relevant to specific actor or regulator sub-types (Figure 1).

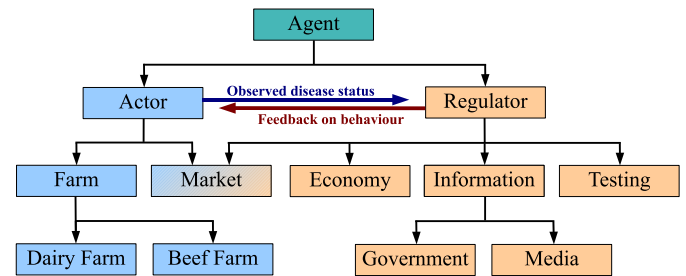


Figure 1. Diagram showing inheritance relationships, and interactions between actors and regulators, within a proposed object oriented animal disease surveillance modelling framework.

## Exemplar Methods and Results

The model described was used to assess the effect of farm heterogeneity and behavioural feedback effects on the observed prevalence of infected premises following a sudden disease incursion event. A population of 1000 farms was set up each with 100 animals, a 10% probability of testing for disease at each time step, a 10% proportion of animals tested conditional on this, and test sensitivity and specificity of 50% and 99.9%. Initially, 10 of these actors were infected (1% infected farms), with a 20% on-farm prevalence of disease, and part-way through the simulation an additional 90 farms were infected at a single time step (total 100 = 10% infected farms). This simulation was repeated 1000 times, recording the observed proportion of infected farms at each time step. The exercise was repeated using heterogeneity between farms in the parameter values for number of animals and propensity to test for disease, again using feedback effects on the propensity to test for disease based on the observed proportion of infected farms, and finally using both farm heterogeneity and feedback mechanisms.

The results obtained are shown in Figure 2. Incorporating behavioural feedback effects into the model greatly increased the time between disease incursion (time 100) and the observed increase in disease prevalence. Including heterogeneity in the model reduced the mean observed prevalence.

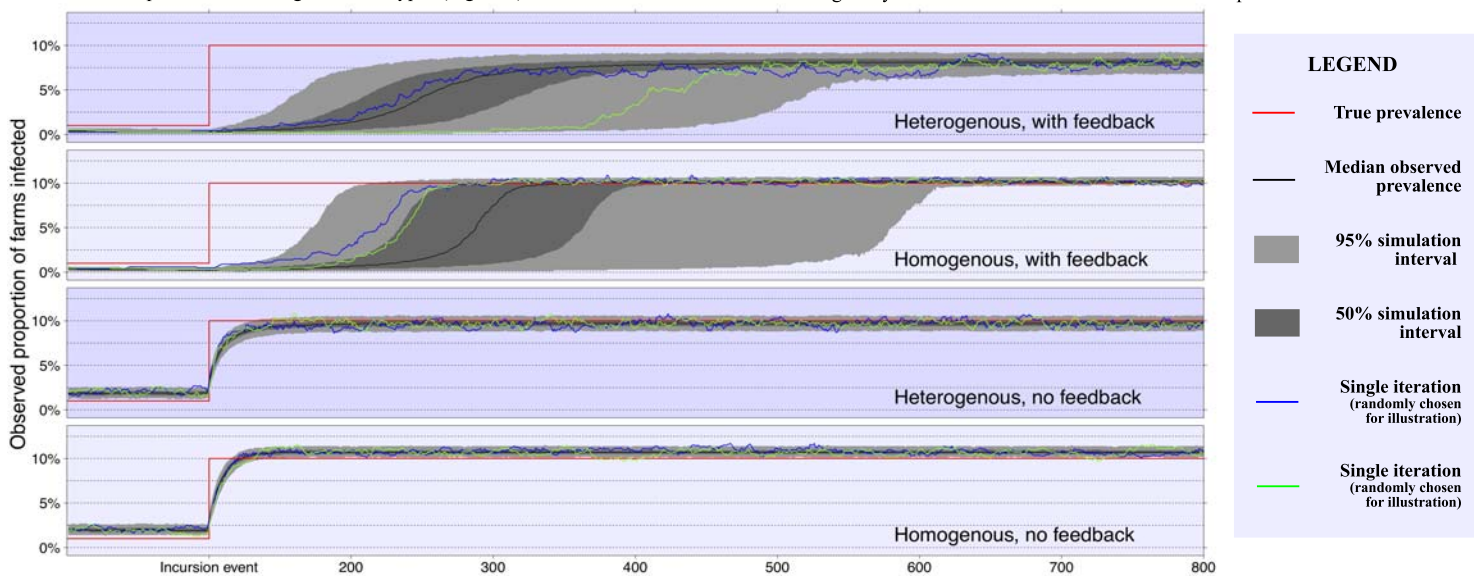


Figure 2. The effect of incorporating farm heterogeneity or behavioural feedback mechanisms on the observed prevalence of infected farms after a disease incursion. Obtained from 1000 simulation iterations based on a population of 1000 farms, with an increase in prevalence from 1% to 10% of infected farms at time point 100. Median, 50% and 95% simulation interval shown alongside two individual iterations.

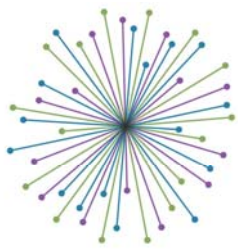
## Discussion

The generic model results presented are focussed on the effects of behavioural feedback mechanisms, and as such are deliberately presented without reference to a specific disease, and with no disease transmission model. Further work is clearly required to adapt the model to any given disease and surveillance system before more focussed recommendations can be made. Nevertheless, it can be seen that incorporating behavioural feedback mechanisms has a profound impact on the observed disease prevalence, especially when also modelling heterogeneity between farms. These effects appear to be non-random, and unpredictable in that this bias changes with heterogeneity and true prevalence. Sequential prevalence estimates are also highly autocorrelated, which introduces a random-walk-like effect in the observed prevalence, when the true prevalence is in fact static. This potential for observed disease prevalence estimates to be inconsistently and unpredictably biased clearly has the potential to affect the performance of disease surveillance strategies. Any possible behavioural feedback mechanisms should consequently be considered and evaluated for future disease surveillance programmes.

The framework presented has a variety of potential uses in disease surveillance scenarios, as well as other applications in which behavioural influences may be important. In the hope that the model might be used and developed in the wider community, we have set up a shared development environment for the model framework, using freely available and open source revision control software. Any researchers interested in collaborating on this project are invited to contact the corresponding author.

## Conclusions

- Behavioural feedback mechanisms can have a dramatic effect on the observed disease prevalence, and compound the difficulties caused by heterogeneity between farms.
- Our agent-based approach is able to account explicitly for feedback mechanisms, as well as farm heterogeneity and imperfect diagnostic tests.
- Application of this method to specific population and disease parameters will provide more accurate evaluation of disease surveillance strategies.



# The systematic review process: comparing databases for pet population searches



The University of Nottingham

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**CENTRE FOR EVIDENCE-BASED VETERINARY MEDICINE**  
Putting research into practice

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## Background:

A systematic review aims to provide an exhaustive summary of literature (published and grey) relevant to a research question. The first step is a thorough search of the databases and citation indexes that are relevant to the topic area. Most databases currently use aids for indexing and retrieving publications. Subject headings (SH) are controlled terms or descriptors that are manually assigned to publications by the database indexers or librarians. They consist of a set of terms or descriptors used in a hierarchical structure which allows searching at various levels of specificity. The databases used in this review each have their own thesaurus of terms: Cab Abstracts™ uses the CAB Thesaurus, EMBASE™ uses a thesaurus known as Emtree® and both PubMed® and Medline® use the MeSH® (Medical Subject Headings) terms thesaurus formulated by the National Library of Medicine's (NLM) library. While these databases are used extensively in systematic reviews, little has been done to demonstrate the difference between subject heading and keyword searching. Currently a systematic review is being undertaken to determine the best methods of determine the demographics and population size of the dog and cat population in the United Kingdom; we use this as a case study for this poster. The aim of the poster is to compare searching with keywords and subject headings using various publication databases.

## Methods:

Medline, CAB and EMBASE were accessed through the Ovid SP interface, whereas PubMed was accessed through the online interface provided by The National Center for Biotechnology Information. Search terms were entered into the respective databases for keyword searches and were as follows: population, population density, population dynamics, demography and census. The subject headings that were suggested by the database search engines were used as comparators for each of the keywords. The results were then tabulated in MS Excel™ and VennGenerator© was used to create Venn diagrams that were proportionally representative of the results, showing overlapping representation between some of the different search terms.

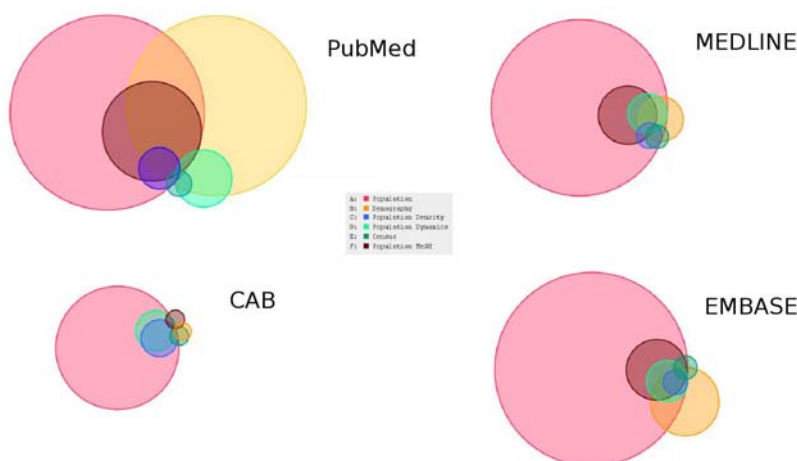


**Table 1. The number of publications found and the proportional differences between keywords and subject headings (SH), when using databases to search for publications for a systematic review.**

	MEDLINE				PubMed				CAB				EMBASE			
	Keyword	Subject headings	Proportion of SH not in Keyword	Proportion of keywords not in SH	Keyword	Subject headings	Proportion of SH not in Keyword	Proportion of keywords not in SH	Keyword	Subject headings	Proportion of SH not in Keyword	Proportion of keywords not in SH	Keyword	Subject headings	Proportion of SH not in Keyword	Proportion of keywords not in SH
Population	750292	83458	0	0.89	921883	238855	0	0.74	367100	8196°	0.33	0.99	908272	86129	0	0.91
Population Density	15989	13201	0	0.17	41614	13187	0	0.68	32230	23185	0	0.28	14692	15861	0.23	0.16
Population Dynamics	38071	56269	0.37	0.06	75637	56302	0	0.26	36798	40221	0.22	0.15	40695	41079	0.06	0.05
Demography	49034	773116	0.94	0.03	791645	772905	0	0.02	7084	4859	0	0.31	114224	112669	0	0.02
Census	12971	2146*	0.34	0.88	15591	2141	0	0.86	8644	2346*	0.31	0.79	13627	51799^	14.02	0.75
All Together	772369	1637247	0.82	0.62	1462966	910909	0	0.38	376161	73644	0.07	0.82	978801	253407	0.01	0.74
Animal Population Groups	1970	833720	0	0.99	836539	83299	0	0.01								

\*Censuses was the subject heading used  
°Populations was the subject heading used  
^Population research was the subject heading used

Figure 1. Venn diagrams showing a proportional estimation of the overlap between the different search terms and subject headings



## Discussion:

The results show that Pubmed has the largest database of information. Both PubMed and the CAB abstracts indexing systems seem to be more accurate with both of them having the lower proportions of publications indexed outside of the keyword searches, with PubMed having all the SH publications checked in this study included in the keyword searches. Some of the databases have large discrepancies between which publications are indexed to a SH when compared with the keyword searches. What is of concern is that both PubMed and Medline are products of NLM and between them there seems to be substantial differences in the way that publications are indexed. This study shows that great care is required when using SH searches as there can be huge variations between the SH and the keyword searches and publications are open to different librarian interpretations when been mapped to a SH. It also shows that if you want a more sensitive search strategy it is better to use both methods of searching whereas SH searches tend to be more specific.

## Acknowledgment

Thanks very much to The University of Nottingham and Novartis Animal Health for sponsoring this research

# Scaling from challenge experiments to the field: predicted impact of vaccination on the transmission of bluetongue virus serotype 8

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## Introduction

- ❖ Bluetongue (BT) is an economically important disease of ruminants caused by bluetongue virus (BTV) and transmitted by *Culicoides* biting midges.
- ❖ The most practical and effective way to protect susceptible animals against BTV infection is vaccination.
- ❖ We investigated the effects of vaccination on transmission of BTV serotype 8 (BTV-8) within and between farms, based on data from challenge experiments generated by Intervet/Schering Plough Animal Health:
  - the basic reproduction number ( $R_0$ ) was used to assess the impact of vaccination within a farm; and
  - a stochastic spatial model for the spread of BTV in Great Britain (GB) was used to assess the impact of vaccination on transmission between farms.
- ❖ Both analyses allow for uncertainty in the parameters related to BTV transmission.

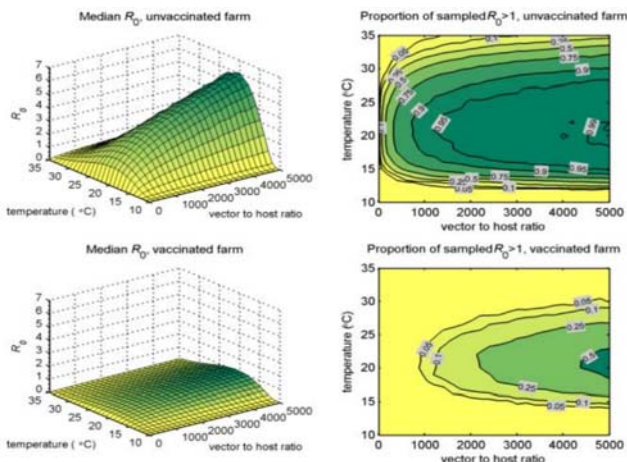
## Vaccine parameters

- ❖ Estimates (95% credible limits) for the vaccine parameters are:

parameter	cattle	sheep
vaccine efficacy (%)	75 (60-85)	98 (89-100)
reduction in duration of viraemia (%)	53 (31-66)	not estimated

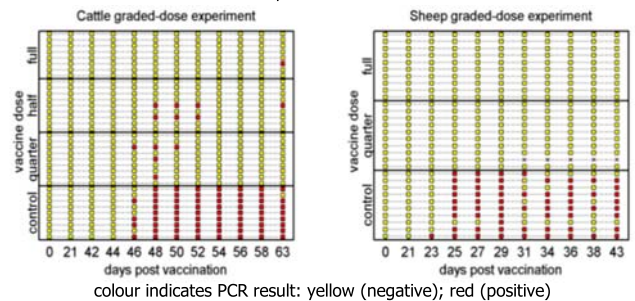
## Basic reproduction number

- ❖ A temperature dependent model was used to compute the basic reproduction number ( $R_0$ ) for BTV [1,2].
- ❖ Replicated Latin hypercube sampling was used to allow for uncertainty in model parameters [1]. Plausible ranges for each parameter were obtained from the literature [1], while vaccine parameters were derived from the challenge experiments.
- ❖ Comparison of  $R_0$  in unvaccinated and vaccinated populations shows that the reduction in  $R_0$  due to vaccination depends only on vaccine parameters.
- ❖ Based on the estimates obtained from the challenge studies, this reduction in cattle is 83% (95% CI:71-90%); in sheep it is 99.8% (95% CI: 98.9-100%).
- ❖ Allowing for uncertainty in the model parameters,  $R_0$  exceeds one in an unvaccinated population at all but the lowest vector-to-host ratios and coolest and warmest temperatures.
- ❖ Vaccination reduces  $R_0$  below one at all but the highest vector-to-host ratios and optimal temperatures; this is robust to uncertainty in the parameters.



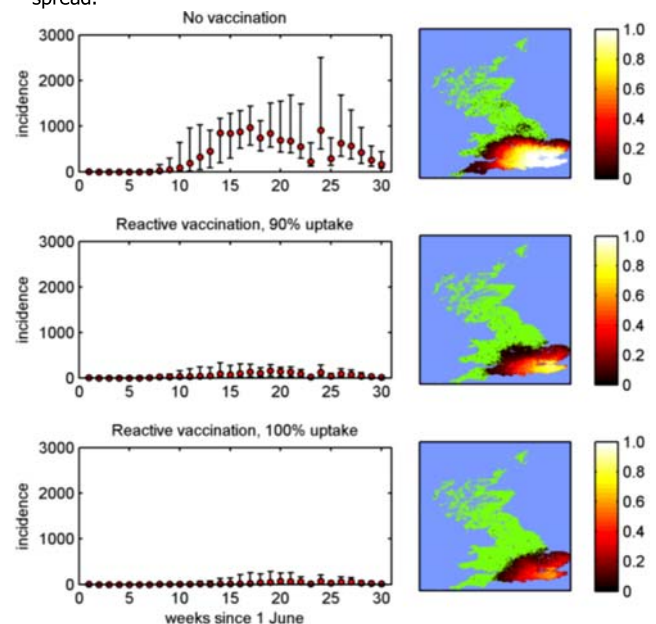
## Challenge experiments

- ❖ Cattle: 35 Holstein dairy calves allotted to four groups of eight or nine animals. Three groups of calves were vaccinated twice (at day 0 and 21) with Bovilis BT8<sup>®</sup> using a full, half or quarter dose. A fourth group served as saline injected controls. Animals were challenged on day 42.
- ❖ Sheep: 30 sheep were allotted to three groups of ten sheep. Two groups were vaccinated at day 0 with a full or quarter dose; the third group served as saline injected controls. Animals were challenged on day 21.
- ❖ Blood samples were taken at regular intervals and the presence of virus was assessed using RT-PCR.
- ❖ The outcomes of the challenge experiments were used to estimate the vaccine efficacy (proportion of animals protected) and the reduction in the duration of viraemia in infected, vaccinated animals.



## Transmission between farms

- ❖ A stochastic, spatially explicit model was used to describe spread between farms in GB and the impact of vaccination [3,4].
- ❖ Three vaccination strategies were considered:
  - no vaccination
  - vaccinate 90% farms in a 50km zone around an infected premises (IP)
  - vaccinate 100% farms in a 50km zone around an IP
- ❖ Vaccination significantly reduced the incidence of disease and spatial spread.



## Conclusions

- ❖ The proportional reduction in  $R_0$  due to vaccination depends only on vaccine parameters and so will be independent of local conditions.
- ❖ From the results of the challenge experiments this reduction is predicted to be 83% and, based on parameters derived to reflect BTV-8 epidemiology in northern Europe, this is likely to be sufficient to reduce  $R_0$  to below one.
- ❖ Vaccination was predicted to reduce significantly the incidence of disease and spatial spread in simulated BTV-8 outbreaks in GB.
- ❖ The methodology used here is ideal for assessing the impact of vaccination on transmission of diseases not amenable to experimental studies (e.g. a vector-borne disease).



# Efficacy of foot-and-mouth disease emergency vaccination: A meta-analysis

Tariq Halasa<sup>1</sup>, Anette Boklund<sup>1</sup>, Sarah Cox<sup>2</sup>, Claes Enøe<sup>1</sup>

## Objective:

A meta-analysis study was conducted to provide a comprehensive assessment of the efficacy of foot and mouth disease (FMD) emergency vaccination, based on the available literature, which may facilitate further economic analysis on the use of FMD emergency vaccination.

## Procedures:

### 1- Inclusion criteria:

Experimental research and symposium papers published in English and unpublished data were considered. Each experiment must have 2 groups: challenged vaccinated (cases), and challenged non-vaccinated (controls).

### 2- Outcome parameter (relative risk-RR):

- a. Clinical protection (absence of clinical disease):

$$RR = \frac{\text{Incidence of clinical disease in cases}}{\text{Incidence of clinical disease in controls}}$$

- b. Virological protection (absence of virus shedding)

$$RR = \frac{\text{Incidence of infection in cases}}{\text{Incidence of infection in controls}}$$

### 3- Meta-analysis procedure:

The RRs were pooled over studies; separately per protection parameter and animal species and classified on virus serotype, using a random effects model. Meta-regression and tests for publication bias were carried out.

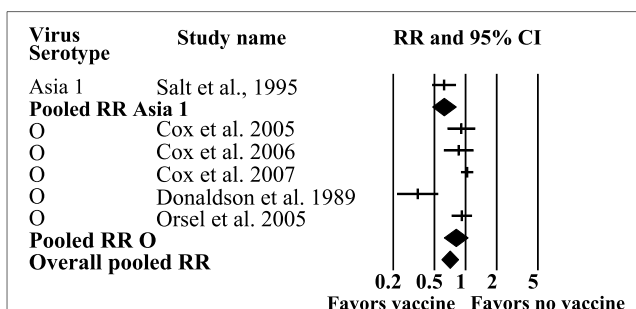


Fig. 1. Forest plot of the relative risk (RR) with the 95% confidence interval (CI) per study, the pooled RR per virus serotype and the overall pooled RR for the virological protection against FMD in cattle following vaccination.

## Results:

Ninety six studies were identified, of which 27 were included in the analysis. Table 1 shows the vaccine efficacy per animal species and protection parameter, e.g. vaccinated cattle have on average 0.13 lower chance of developing FMD clinical signs compared to non-vaccinated cattle.

Animal species / parameter	Pooled RR and 95% CI
Cattle	
Clinical protection	0.13 (0.09-0.18)
Virological protection	0.71 (0.59-0.85)
Swine	
Clinical protection	0.48 (0.36-0.65)
Virological protection	0.67 (0.51-0.87)
Sheep	
Clinical protection	0.31 (0.18-0.53)
Virological protection	0.59 (0.44-0.80)

Fig. 1 shows that vaccinated cattle are generally protected against FMD infection significantly more than non-vaccinated cattle, but the protection may be serotype dependent. Fig. 2 shows that small studies (with large standard error) may have been published, only because they show large and interesting effects. This is an indication of potential publication bias. However, correcting for this bias would not alter the results significantly.

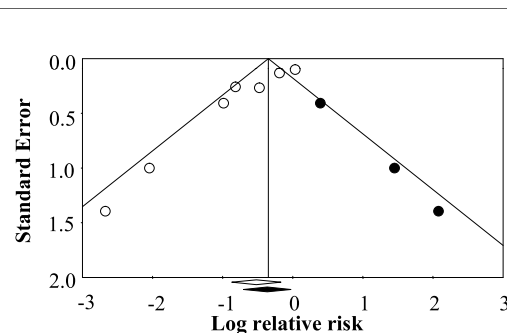


Fig. 2. Funnel plot of the logarithm pooled relative risk of 7 studies (empty circles) quantifying the effect of FMD vaccination against infection in sheep. The dark spots are the potential missing studies (if they had existed, the effect would have shifted from 0.59 (0.4-0.8); the white diamond under the X-axis, to be 0.68 (0.50-0.99); the black diamond under the X-axis, and complete symmetry would have been reached, removing the effect of publication bias).



## Conclusions:

- Emergency vaccination against FMD provided protection against clinical disease and against FMD infection in cattle, swine and sheep.
- No significant publication bias was observed in the corresponding literature.
- The results can be used in simulation modeling to assess the economic consequence of FMD emergency vaccination.



# Incidence and risk factors of Highly Pathogenic Avian Influenza (HPAI) infection in smallholder, village-based duck flocks in Indonesia

Joerg Henning<sup>1</sup>, Hendra Wibawa<sup>1,2,3</sup>, Didik Yulianto<sup>3</sup>, Akhmad Junaidi<sup>3</sup>, Joanne Meers<sup>1</sup>

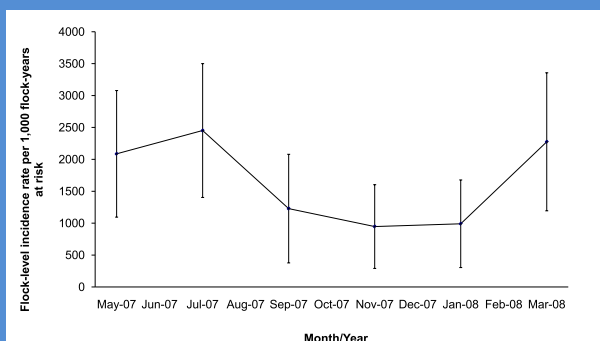
<sup>1</sup>University of Queensland, Australia; <sup>2</sup>Australian Animal Health Laboratory, Geelong, Australia; <sup>3</sup>Disease Investigation Centre Wates, Indonesia

**Background:** All published risk factor studies from countries where H5N1 HPAI is widespread are based on disease outbreak occurrence rather than on incidence of infection, because the latter requires undoubtedly a more intense and costly monitoring of poultry populations for their infection status. However, as not all infections of ducks result in apparent signs of disease, risks associated with the management of ducks that result in infection of flocks might differ from those risks resulting in outbreaks. Therefore we investigated HPAI incidence of duck flocks and identified risk factors for new H5 HPAI virus introductions into “stationary”, village-based duck flocks.

**Methods:** Ducks on 96 small-holder duck farms in 4 districts (Sleman, Magelang, Bantul, Kulon Progo) of central Java, Indonesia, were surveyed once every two months over 12 months in 2007/2008. Sera were tested for H5 antibodies using the haemagglutination inhibition test. Questionnaire surveys were conducted to record information on the management of flocks in-between visits (i.e. the risk factors). We used in a logistic GEE model with an exchangeable correlation structure to identify risk factors associated with incidence of H5 antibody positivity of flocks.



## Results:



Explanatory variable	Category	Farm status		OR (95%CI)	P
		H5 negative (%)	H5 positive (%)		
Carcasses of dead birds eaten	No	233 (76.6)	2 (33.3)	10.2 (1.2-85.9)	0.03
	Yes	71 (23.4)	4 (66.7)		
Ducks roaming throughout the village	No	219 (77.1)	16 (61.5)	2.8 (1.2-6.9)	0.02
	Yes	65 (22.9)	10 (38.5)		
Duck confinement over night on the farm	No	36 (65.5)	199 (78.0)	0.4 (0.2-0.9)	0.04
	Yes	19 (34.5)	56 (22.0)		
Sudden deaths of birds	No	212 (74.4)	23 (92.0)	0.2 (0.1-0.8)	0.02
	Yes	73 (25.6)	2 (8.0)		

Flock-level incidence rate was high at the first flock-sampling and peaked in July 2007 and then decreased and remained low until January 2008, before it rose again in February 2008. The scavenging and roaming of ducks through the village increased the risk of duck flocks developing H5 antibodies, but also the consumption by the farmer of carcasses of dead or sick birds increased the risk (although this was only conducted on six out of 310 farm-samplings over the study period). If ducks were housed overnight on the farm, the risk of flocks developing H5 antibodies was reduced and surprisingly, if dead birds were found on the farm in the preceding two months before the flock-sampling, the risk of flocks developing H5 antibodies was also reduced.

**Discussion:** The carcasses of sick birds potentially contain a high titre of virus. Through the process of slaughtering sick birds for consumption or the disposal of the remains, the virus could be spread and might result in the infection of ducks on the farm. The roaming of ducks through the village allowed contact with various other birds, people and other possible sources of infection. If birds were confined overnight on the farm, the risk of infection was reduced, most likely because less contact could be made with potentially infected animals. It seems that sudden deaths made farmers aware of the bio-security risk these carcasses pose, hence farmers responded quickly to stop the spread of infection from these carcasses by immediately removing and disposing of the bodies.

# The role of rice paddy owners, transporters of ducks and hatcheries in influencing the spread of Highly Pathogenic Avian Influenza (HPAI) virus in Indonesia

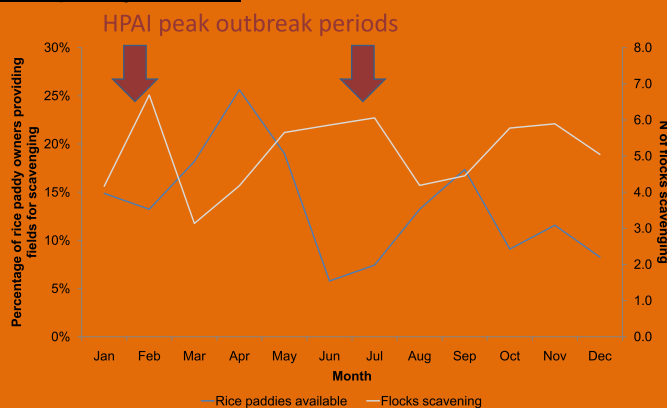
Joerg Henning<sup>1</sup>, Waluyo Priyono<sup>2</sup>, Didik Yulianto<sup>2</sup>, Akhmad Junaidi<sup>2</sup>, Joanne Meers<sup>1</sup>  
<sup>1</sup>University of Queensland, Australia; <sup>2</sup>Disease Investigation Centre Wates, Indonesia

**Background:** Previous investigation focussed on identifying hazards associated with the management of duck flocks, that might increase the risk of HPAI spread. However, various other professions are associated with the duck production system, directly or indirectly. These include transporters of moving duck flocks, rice paddy owners who offer their rice fields for scavenging, and hatcheries which provide ducklings and/or purchase eggs from duck flocks. Their management practices might be related to external, biosecurity hazards of HPAI spread.

**Methods:** Rice paddy owners (N=121), transporters of moving duck flocks (N=30) and hatchery owners (N=75) were interviewed in 2009 in central Java, Indonesia, using cross-sectional questionnaire surveys.

## Results:

### Rice paddy owners



The majority of rice paddy farmers provided their paddies for scavenging in the period from March to May with a peak in April and then again August to October with a peak in September. The number of flocks scavenging per paddy per month varied between 3-4 flocks in the peak scavenging months and 5-6 flocks in the months when less paddies are available. Usually farmers did not receive any payment from the duck owners. The most important benefit for paddy farmers was the control of snails (69%), followed by duck faeces providing fertiliser (43%). The majority of duck owners disposed carcasses of ducks that died during scavenging by burial (50%), but many owners just threw them into nearby rivers (38%).

### Transporters of duck flocks

About 90% of transporters combined flocks from different farms into a one load with a median number of 14 duck farms visited to obtain one load. 67% of transporters indicated that ducks had contact with each other during transport. A median number of 16 journeys were conducted per year to scavenging locations (median distance travelled 90km). About 65% of transporters also transported duck feed, 14% chickens, 25% other birds, 25% other animals than birds and 39% eggs together with ducks on the same load. The most common deaths during transport were from physical injuries, followed by dehydration or diseases. About 57% of transporters threw ducks that died during transport into rivers or paddies. All transporters removed faeces from the vehicles, but only 13% used disinfectant on the vehicle surfaces.



### Hatcheries

About 49% of hatcheries produced their own eggs, 52% purchased eggs from stationary and 25% from moving duck flocks and 18% obtained eggs from professional egg traders. The majority of hatcheries used modern egg incubators (58%), but the use of traditional methods (e.g. oil lamps) (34%) or muscovy ducks (18%) was also common. In close distance to 56% hatcheries, chickens or ducks were kept, most of them free-ranging. Although duckling pens were cleaned in 33% of hatcheries, disinfection was only conducted in 12% of hatcheries.

**Discussion:** HPAI can only be successfully controlled when the complexity of duck production, with all its facets, is understood. We provided new insights into HPAI epidemiology by identifying potential HPAI hazards related to activities not conducted by duck farmers, but by their associates. This can provide the basis for recommendations for effective HPAI prevention, before the virus is entering susceptible duck flock populations.

# Understanding the management of smallholder, village-based duck flocks in Indonesia – opportunities for Highly Pathogenic Avian Influenza (HPAI) control

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<sup>1</sup>University of Queensland, Australia; <sup>2</sup>Australian Animal Health Laboratory, Geelong, Australia; <sup>3</sup>Disease Investigation Centre Wates, Indonesia

**Background:** High duck numbers are considered a major risk factor for HPAI outbreaks in Thailand, Vietnam and Indonesia (Gilbert et al., 2008), but detailed information on duck management factors that potentially influence the risk of outbreaks and HPAI virus prevalence is still lacking. We describe here the management of smallholder duck flocks with the aim to highlight possible hazards associated with this farming system.

**Methods:** A cross-sectional questionnaire survey was conducted on 96 smallholder stationary duck farms in four districts in central Java, Indonesia (Sleman, Magelang, Bantul, and Kulon Progo). These farms were monitored in a longitudinal study to describe temporal patterns of H5 infection in unvaccinated scavenging ducks and chickens (Henning et al., 2010). Questionnaires were used to record information on flock management, feeding, housing, scavenging practices, health status of ducks and mortalities due to HPAI and other diseases. The questionnaire contained 64 questions, comprising multiple choice questions, yes/no questions and open-ended questions



## Results:

Identified hazards	Potential impacts
Intermingling between ducks and chickens on the same farm is common (48% of farms)	Enhancing interspecies HPAI virus transmission on the same farm
Frequent contact between ducks and neighbours' chickens (44% of farms)	Enhancing interspecies HPAI virus transmission within the village
Ducks often disperse on the way to the scavenging locations (33% of farms)	Enhancing excreted HPAI virus spread on transport routes
Frequent visits of the same rice paddies by various duck flocks are common (88% of farms)	Increasing the excreted HPAI virus concentration in the scavenging locations and enhancing contacts between duck flocks
During scavenging frequent contact of ducks with other ducks, chickens, people, wild birds occurs (88%, 30%, 80% and 77% of farms, respectively)	Enhancing interspecies HPAI virus transmission in scavenging locations
Singing birds often kept as pets on duck farms (17% of farms)	Enhancing interspecies HPAI virus transmission on the same farm
Main predators visiting the scavenging areas are Garangan (Small Asian Mongoose) and feral cats (18% of farms)	Predators are susceptible to HPAI infection and might also play a role in the spread of the HPAI virus
Although the majority of duck owners uses rice paddies for scavenging throughout the year (69% of farms), almost a quarter of owners use them only seasonally	Might be related to the seasonal occurrence of HPAI outbreaks
Use of droppings as fertilizer is very important or important for almost 60% of duck farm owners	Highlights the integrated crop-harvest-duck production practiced by small-scale ducks owners and enhances HPAI virus spread in the environment
Burning of litter from the enclosures is uncommon (2% of duck farms)	Enhances HPAI virus concentration in the environment
Most important duck health problems specified: deaths from pesticides in the rice paddies, problems that inhibit the ability of ducks to scavenge (e.g. leg injuries) and external parasites	Avian influenza is not considered to be of high importance for duck owners; hence vaccination of ducks against HPAI or preventive culling of infected ducks during outbreaks was not conducted on study farms

**Conclusion:** We were able to summarize important hazards associated with the management of smallholder duck flocks that could potentially promote HPAI spread. These hazards should be the focus of strategies to reduce the risk of HPAI transmission. The identification of risk factors associated with HPAI outbreak occurrence or infection was not the aim of this research, but with this descriptive survey we were able to provide scientifically valid data for HPAI risk assessments or other HPAI risk-based investigations.

## References:

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 Henning J, Wibawa H, Morton J, Usman TB, Junaidi A, Meers J, 2010. Scavenging Ducks and Transmission of Highly Pathogenic Avian Influenza, Java Indonesia. *Emerg Infect Dis*, 16:1244-50.



# Reduction of *Coxiella burnetii* shedding by vaccination



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## Introduction

Human Q fever cases in the Netherlands increased dramatically between 2007 and 2009. In response to this increase, dairy goats and dairy sheep in the high incidence area were vaccinated with Coxevac®. Despite vaccination, all pregnant dairy goats and dairy sheep in herds positive for Q fever were culled early 2010, thereby precluding any field analysis of vaccine efficacy. However, there was an opportunity to sample animals shortly after they were humanely killed.

## Goal

The purpose of this study was to quantify the effect of vaccination on bacterial load in excreta of pregnant goats and sheep.



## Materials & Methods

On the day of culling, samples of uterine fluid, vaginal mucus, and milk were obtained from 957 dead pregnant animals of 13 herds. Per herd, ~50 pregnant and lactating animals (old animals), and ~50 nulliparous animals (young animals) were sampled. Quantitative real-time PCR was performed for all samples. Results for the 3 sample types were cycle threshold ( $C_t$ ) values, in which a lower value indicates a higher bacterial load. All samples with  $C_t < 40$  were considered as positive. A negative result indicated that no specific signal was detected in a maximum of 40 cycles.



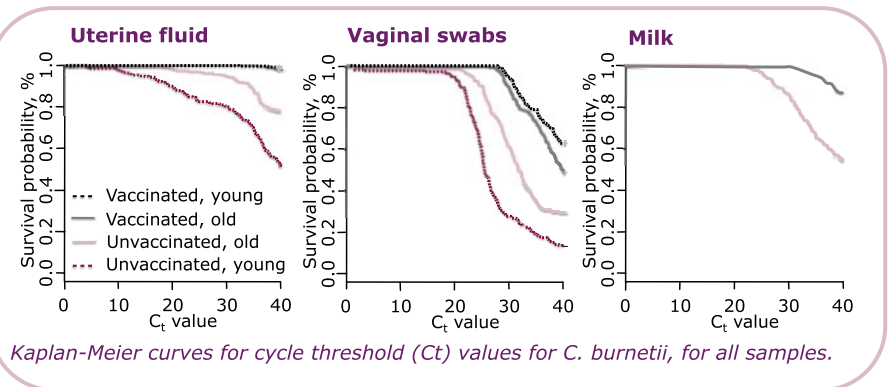
## Results

Prevalence and bacterial load were reduced in animals from vaccinated herds compared with animals from unvaccinated herds. This effect was most pronounced in animals during their first pregnancy.

The bacterial loads ( $C_t$  values) in uterine fluid, vaginal swabs and milk are presented in the figure below. Vaccinated animals hardly have any positive tests results in uterine fluid. In vaginal swabs and milk, vaccinated animals are less likely to test positive than unvaccinated animals, and when they do, they have higher  $C_t$  values (~ lower bacterial loads).

In the tables below, the results of multivariate logistic regression and univariate survival analysis are presented for uterine fluid. In uterine fluid, vaccinated animals had a very low odds ratio (OR) for testing positive and had a hazard ratio (HR) that was half that of unvaccinated animals, which indicated that unvaccinated *C. burnetii*-positive animals had higher relative amounts of bacteria on the basis of  $C_t$  value. These effects were similar for vaginal mucus and milk.

## Quantitative PCR results



## Prevalence

Group	OR (95% CI)
Unvaccinated, young	1
Unvaccinated, old	0.44 (0.25–0.78)
Vaccinated, young	0.005 (0.0002–0.12)
Vaccinated, old	0.03 (0.002–0.58)

*Multivariate logistic regression of prevalence of Coxiella burnetii in uterine fluid. A random herd effect was included.*

## Conclusions

This observational study showed reduced prevalence and bacterial load in uterine fluid, vaginal swabs and milk from vaccinated animals, in comparison to unvaccinated animals. These effects were most pronounced in animals during their first pregnancy. Results indicate that vaccination may reduce bacterial load in the environment and human exposure to *C. burnetii*.

## Bacterial load

Group	HR (95% CI)
Unvaccinated	1
Vaccinated	0.49 (0.39 – 0.70)

*Univariate survival analysis of PCR  $C_t$  values for Coxiella burnetii in positive uterine fluid samples ( $C_t \leq 40$ )*

# A COHORT STUDY TO INVESTIGATE THE ASSOCIATION BETWEEN EWE MILK SOMATIC CELL COUNT AND LAMB WEIGHT

Huntley, S., Cooper, S., Green, L.E.

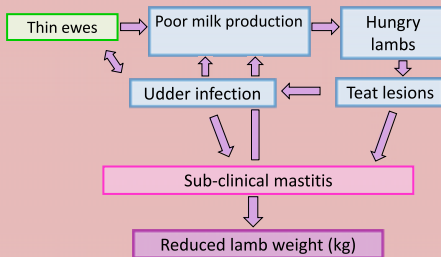
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## Introduction

Subclinical mastitis results in a reduced milk yield in dairy ewes<sup>1</sup> and in decreased weight of lambs reared by ewes<sup>2,3</sup>. Previous studies on the effect of subclinical mastitis on lamb growth have not investigated and accounted for the complex associations of other variables on the longitudinal relationship between subclinical mastitis and lamb weight.

## Hypotheses

- A change in lamb weight is associated with
- Somatic cell count (SCC)-subclinical mastitis
- Teat lesions-external damage



## Methods

**Milk sample and data collection** : One farm, January to May 2010.

Data collected:	Ewes (n=67)	Lambs (n=101)
at each observation:	SCC of milk from each udder half	Weight
	Body Condition Score (BCS)	Diarrhoea
	Teat damage	Orf
	Abnormalities of the udder or milk	
at lambing only	Litter size	sex

**Cohorts followed**

Cohort	Breed	Age	Number of lambs
1 (n=16)	Suffolk mules	2 and 6 yrs	1
2 (n=22)	Suffolk mules	2 and 6 yrs	≥2
3 (n=17)	North of England mules	9 yrs	1
4 (n=12)	North of England mules	9 yrs	≥2

**Data analysis using multilevel linear regression models**

MLwiN 2.11 (Centre for Multilevel modelling, University of Bristol)

**Lamb weight model:**

Dependent variable: lamb weight (kg)

Random effects: ewe, lamb and observation as levels 3, 2 and 1

**Somatic cell count model:**

Dependent variable: log<sub>10</sub> SCC (cells/ml)

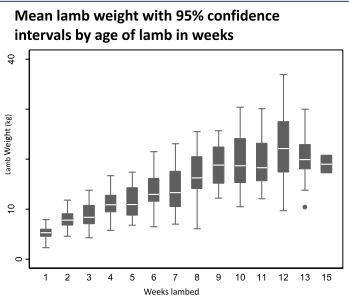
Random effects: ewe, udder-half and observation as levels 3, 2 and 1

## Results

### Data summary

Variable	ewes (n=67)	Variable	Lambs (n=101)
Rearing singles	35	Female	42
Rearing twins	31	Male	59
Rearing triplets	1	Orf (at any observation)	16
≥ 1 teat lesion (at any observation)	41	Diarrhoea (at any observation)	25

## Lamb weight



### Lamb weight model

After accounting for lamb age, birth weight, litter size, whether the lamb had diarrhoea, and supplementary concentrate fed to the ewe before lambing, lower lamb weights were associated with:

**higher ewe mean SCC**

Lambs reared by ewes with a mean log<sub>10</sub> SCC of above 5.6 (400,000 cells per ml) weighed on average 1.4kg [-2.1,-0.7] less at that observation.

**the observation of a new teat lesion**

Lambs reared by ewes on which a "non-traumatic" teat lesion (warts, spots, orf) was at that visit weighed 0.5kg less [-1.0, 0.1] (significant at 90%)<sup>†</sup>.

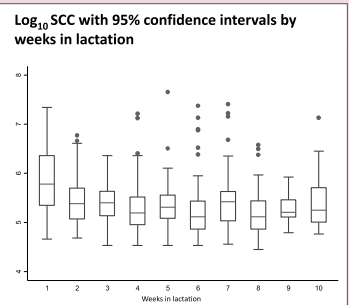
Lambs reared by ewe on which a "traumatic teat lesion" (bites tears and chapping) was observed at the previous visit weighed 0.9kg less [-1.4, -0.1].

**teat position**

The teat position associated with heavier lambs was at 4 and 8 o'clock. Lambs reared by ewes with the most medial teat positions weighed on average 1.4kg less [-2.5,-0.3] and those in the more lateral teat positions weighed up to 1.5kg [-2.6, -0.4] less than those reared by ewes with teats in the 4 or 8 o'clock position.

Lambs reared by ewes that were 9 years old weighed on average 2.4 kg less [-3.4,-1.5] at each observation than those reared by ewes that were 6 years old. Lamb gender, ewe body condition, breed and udder size were non significant variables.

## Somatic cell count



### Somatic cell count model

After accounting for days in lactation and average lamb weight, SCC was significantly higher in:

**ewes with a lower udder drop**

For every increase of 1 cm in udder drop there was an associated 0.04 increase in log<sub>10</sub> SCC [0.01, 0.07] (where "lower" is a greater distance (in cm) as measured from base to the apex at the bifurcation of the udder with the ewe standing).

**ewes with a greater cross-sectional area of both teats**

For every 1 cm<sup>2</sup> increase in total teat cross-sectional area, there was an associated 0.03 [0.01, 0.05] increase in log<sub>10</sub> SCC.

**old and thin ewes**

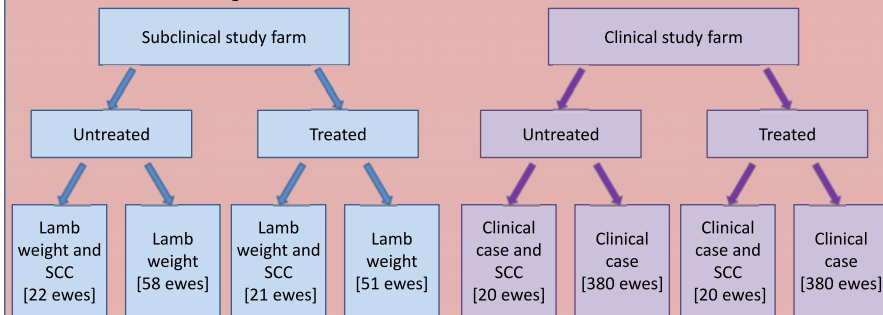
9 year old ewes of BCS<3 in lactation and 6 year old ewes in BCS of <2 in lactation had significantly higher SCCs when compared to young fit ewes (2 years old and of BCS>3). For example, the thinnest 9 year old ewes of BCS>2 had a 0.27 higher log<sub>10</sub> mean SCC and thinnest 6 year old ewes had an increase of 0.70 0.27 higher log<sub>10</sub> mean SCC [0.23,1.17].

## Conclusions

After controlling for ewe and lamb variables, lower lamb weight can be explained by subclinical udder infection of the ewe and the occurrence of teat lesions. There is an association between udder conformation and the level of SCC and lamb growth which warrants further investigation. By improving knowledge of the dynamics of SCC, we provide further understanding of how to maintain udder health of ewes in order to benefit to farmers with ewes rearing lambs for the meat industry.

## Further work

Intervention study to assess the effect of a broad spectrum intramammary "dry off" antibiotic at weaning on subclinical and clinical mastitis.



## Acknowledgements

We would like to thank the farmer of the study farm, Quality Milk Management Services Ltd, staff and colleagues at the University of Warwick for their assistance and the English Beef and Lamb Executive for funding the study.

# Adenovirus type 3 infections in camels in Sudan

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## Introduction

Galbreath *et al.* (1994) reported the isolation of adenovirus from lungs of young llamas with pneumonia and hepatitis. Serologically antibodies against adenovirus were detected in 1.3% of dromedaries in Nigeria (Olaleye *et al.*, 1989), in 93% of 270 llamas in USA (Picton, 1993) and in 43 of 120 camels in Egypt (Hadia *et al.*, 2001).



## Materials and methods

### Samples collection:

A total of 239 pneumonic lung specimens and 260 camel sera were collected at four different localities in Sudan.

### Sandwich ELISA for adenovirus 3 antigen and antibody detection:

Sandwich ELISA kits for adenovirus 3 antigen and antibody detection (Bio-X Diagnostics, Jemelle, Belgium) were used according to the instructions of the manufacturer.

## Results

### Adenovirus type 3 antigen detection:

Using sandwich ELISA, 3 out of 239 pneumonic camel lung specimens were found positive (1.3%), the details are presented in Figure 1.

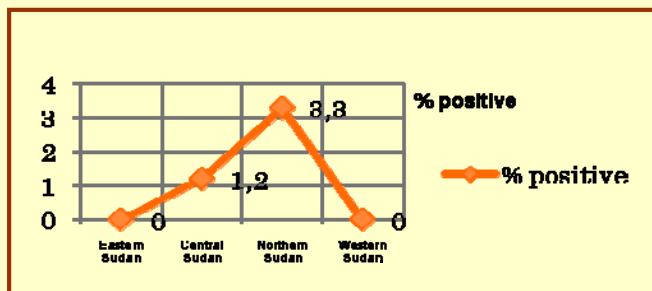


Figure 1: Detection of adenovirus type 3 antigen in camel lungs in different localities in Sudan using ELISA (2000-2006)

### Adenovirus type 3 antibody detection:

Indirect ELISA kits for detection of antibodies to adenovirus 3 were applied on 260 camel sera with 90% positivity (Figure 2).

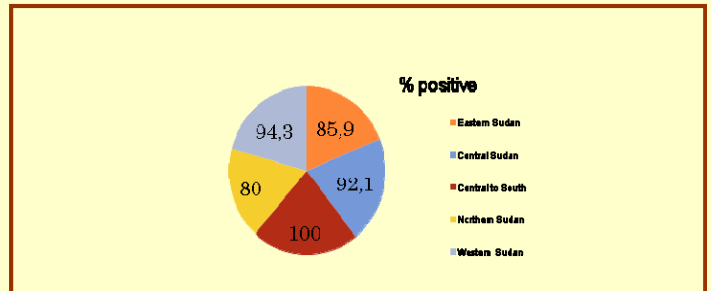


Figure 2: Detection of antibodies to adenovirus 3

## Discussion

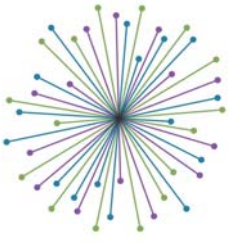
- ✓ In this study, adenovirus antigen was detected in 1.3 % of tested camel lungs using ELISA.
- ✓ This is the first report for adenovirus antigen detection in camels in Sudan and probably elsewhere.
- ✓ This finding supported by the previous reports on the association of adenovirus with pneumonia in camelidae (Galbreath *et al.*, 1994, Mattson, 1994 ).
- ✓ The detected seroprevalence of adenovirus in this study was 90% which is considered high prevalence and is far higher than that reported in previous publications.

## Conclusion

Adenovirus infection in camels of the Sudan is widely distributed. 21.8% of positives showed high titers indicating recent and/or multiple infections.

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CENTRE FOR EVIDENCE-BASED  
VETERINARY MEDICINE  
Putting research into practice

# Capturing patient data in small animal veterinary practice

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## Introduction

The principle of Evidence-based Veterinary Medicine (EVM) is to employ the best relevant, most up-to-date information used by veterinarians in clinical practice to enhance clinical decision making. In order to identify areas in which research is needed, it is necessary to understand which conditions present most commonly, and determine perceived gaps in information. A relatively new methodology which may assist in identifying these gaps in veterinary knowledge is the extraction of clinical data from practice management software (PMS) systems to identify common conditions. This type of methodology has been used effectively in human healthcare; however veterinary medicine has only recently begun to realise its potential.

## Background

Many methods are currently in place to capture emerging or prevalent disease. These systems rely on collecting new data as cases present and assimilating a dataset of diagnoses and presenting complaints. However the increasing reliance on PMS systems to record clinical data means much of this information is already captured in vet PMS systems. Methods that access this information will allow examination of historical data, including treatment success.

Alongside current systems, this new methodology will provide a complete picture of the owned vet visiting companion animal population within the UK and enable epidemiological studies to be performed on a much larger scale.



## Method Development

A pilot study has been designed between the Centre for Evidence based Veterinary Medicine (CEVM) and a commercial PMS system and their clients. This collaboration will allow access to historical data providing an initial clinical portfolio for further method development. This will include retrospective records searching.

The aim of the study will be to evaluate the use of PMS systems for capturing veterinary clinical information and to examine the health of the UK vet visiting companion animal population. Summaries of medical records from real time consultations will be examined in situ and via PMS extraction and compared, to validate the efficiency of the extraction. Once validated the method will allow the extraction of clinical data for veterinary epidemiology and evidence-based research.



## Next Steps

Further work will examine how much information is actually recorded by the veterinarian on the PMS system, a second aim will be an epidemiological study using data extracted from PMS systems to examine the role of clinical nutrition in the management of disease.

## Discussion

It is thought that up to 90% of practices in the UK use a computerised practice management system for their clinical records with many different systems currently available. Developing efficient ways of accessing the PMS data for analysis across different practice systems would enable vast amounts of clinical data to be collated and analysed. Until standard methods of recording data between and across practices are in place, this extraction will be an invaluable tool for evidence-based veterinary research. Potential limitations of the method may include under or over reporting of certain diseases, in particular those that are easily diagnosed may be recorded more frequently<sup>1</sup>.

## Acknowledgement

Thank you to the PMS system provider and their clients for their collaboration and continued support, also to The University of Nottingham and Novartis Animal Health for sponsoring this research.

## References

<sup>1</sup> Elizabeth M. Lund, P. Jane Armstrong, Claudia A. Kirk, Linda M. Kolar, Jeffrey S. Klausner (1999) *JAVMA*, Vol 214, No. 9.



# Use of a two-part model to identify herd- and litter-level factors associated with *Isospora suis* occurrence in Greek finishing-pig herds.

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## Background & Objective

Neonatal coccidiosis, caused by *Isospora suis* (*I.suis*), occurs in all types of farrowing facilities and under all types of management systems (Mundt et al., 2005). In this study we identified herd- and litter-level factors associated with the odds and the level of *I.suis* oocyst excretion in Greek pig herds.

## Farms, Sampling & Testing

Data were collected from 55 farrow-to-finish herds. From each herd and depending on the herd size, at least five litters, in their second week of life, were coprologically sampled. Each litter-sample comprised a pool of individually collected faeces from half of the piglets. Oocyst excretion, expressed as oocysts per gram of faeces (OPG), was determined, by a modified McMaster technique.

## Questionnaire data

A standardized risk-factor questionnaire (available from the first author upon request) was used to collect information on the use of toltrazuril (BAYCOX® 5%) and other herd- and litter-level factors that could be associated with *I. suis* infection.

## Modeling

OPGs are semi-continuous data (Figure 1). Thus, we used a two-part model (Figure 2) to simultaneously identify factors associated with the odds and/or the amount of litter-level oocyst excretion (Liu et al., 2008). Initially, all factors were screened one by one in univariable models. Following a backwards elimination procedure factors significant at  $P < 0.05$  were retained in the final model.

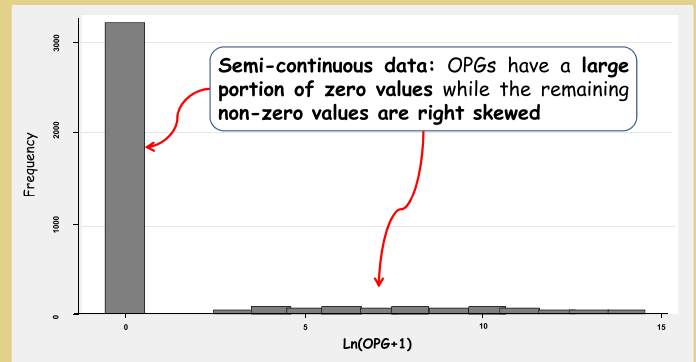
In all models random-effect were incorporated for herd in both parts. Further, a correlation term between the random-effects of the two parts ( $\rho_{CEC}$ ) captured the biologically plausible fact that piglets and litters with higher odds of excretion also tended to excrete more oocysts.

Models were built in the aML software.

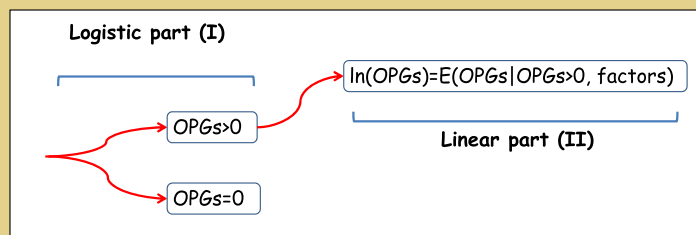
**Table 1. Factors associated with the (I) risk and the (II) level of OPG excretion**

Logistic part (I)		Odds Ratio (95% C.I.)	P
Toltrazuril	Yes	0.2 (0.1 ; 0.5)	0.001
	No	1	
Clean entire farrowing room	Yes	0.3 (0.1 ; 0.9)	0.039
	No	1	
Fostering after 1 <sup>st</sup> day	Yes	4 (1.0 ; 16)	0.045
	No	1	
Metallic pen floor*	Yes	2.5 (1.1 ; 5.7)	0.034
	No	1	
Pens per room*	Continuous	0.93 (0.9 ; 0.97)	0.001
Number of employees	Continuous	0.6 (0.5 ; 0.97)	0.042
Linear part (II)		Coefficients (95% C.I.)	P
Toltrazuril	Yes	-1.8 (-0.6 ; -2.9)	0.004
	No	ref.	
Employees entering in pens	Yes	2.1 (0.6 ; 3.6)	0.009
	No	ref.	
Pens per room*	Continuous	-0.04 (-0.01;-0.08)	0.023
$\rho_{CEC}$		0.99	0.005

**Figure 1. Frequency distribution of the oocysts per gram (OPG) of pooled faeces from litters, in their second week of life, from 55 farrow-to-finish herds**



**Figure 2. The two-part model accounts for the concentration of excessive zero-valued OPGs. It uses logistic regression to predict the probability of occurrence of a non-zero OPG and linear regression to predict the amount of the non-zero OPGs**



## Results

The odds of *I.suis* litter-level excretion were reduced when (i) piglets were treated with toltrazuril, (ii) the farrowing rooms were completely emptied and cleaned after weaning, (iii) piglets were not cross-fostered between litters after the 1st day of life and (iv) the flooring of the pens was plastic. Further, (v) larger farrowing rooms and (vi) more caretakers in the farrowing rooms reduced the odds of excretion.

The level of OPG decreased with (i) toltrazuril-treatment, (ii) larger farrowing rooms and (iii) caretakers avoiding walking into farrowing pens.

## Discussion

Good managerial and hygiene-related practices, which affect between-pen transmission and environmental persistence of the parasite reduce the risk and the amount of oocyst excretion. Hence, control of neonatal coccidiosis can be bettered by the identification and implementation of good-managerial-practice protocols, additionally to toltrazuril treatment.

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# Classical Swine Fever Control

## Oral mass immunisation under uncertain virulence

M. Lange<sup>1\*</sup>, S. Kramer-Schadt<sup>2</sup>, H.-H. Thulke<sup>1</sup>



### Objective

Comparative assessment of adaptive spatial schemes to immunise wild boar populations against Classical Swine Fever (CSF) by oral mass vaccination. A spatially explicit, individual-based, stochastic simulation model was applied. Uncertainty about the severity of the disease outcome was considered.

### Introduction

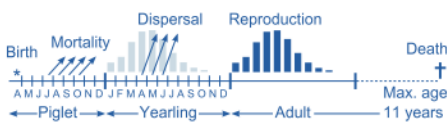
In recent years, Classical Swine Fever Virus (CSFV) circulated in wild boar of several European countries and caused high economic impact when entering livestock. Much effort was spent on oral vaccination campaigns. The effect of the measure on disease dynamics was not fully understood due to variability of disease outcome and the shift of CSFV

to decreased virulence during the last decades. Particularly, efficient spatio-temporal design of vaccination protocols is still debated. Therefore, we assessed spatially adaptive vaccination scenarios with regard to the efficacy in limiting spread and survival of the infection in an infected boar population while considering uncertain virulence.

### Methods

#### The wild boar model

- Raster of home range cells of wild boar groups
- Wild boars as individuals
- Seasonal, age-dependent mortality and fertility



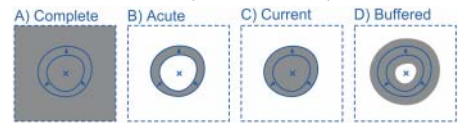
#### The virus model

- Stochastic disease outcome (transient, lethal acute, lethal chronic)
- Transient infections cause an infectious period of one week
- Lethal infections cause exponentially distributed infectious period



#### Simulation experiments

- Simulation of four baiting strategies (A complete, B recent infected area, C cumulative extension [current strategy], D like B but with spatial buffer)
- Simulation of different disease outcome, i.e. case mortality (0 ... 1) and infectious period after lethal infection (1 ... 10 weeks)



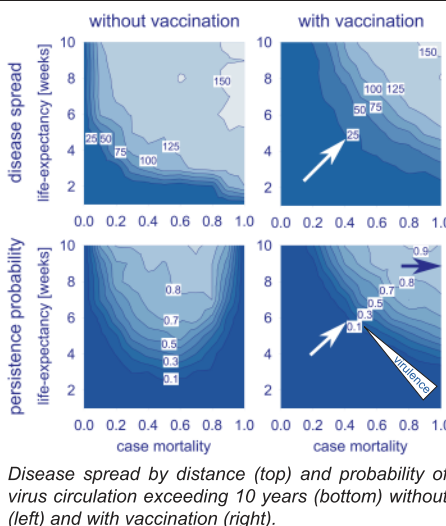
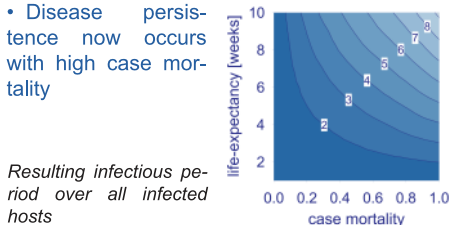
### Results

#### Without vaccination

- Distance spread depends on the mean infectious period over all infected hosts
- Disease persistence depends on the infectious period over all hosts and case mortality

#### With vaccination

- Distance spread only for long infectious period over all infected hosts
- Disease persistence only for long infectious period over all infected hosts
- Disease persistence now occurs with high case mortality



#### Comparison of strategies

- Virtually equal capacity to reduce spread and foster disease eradication by baiting of the **entire landscape** (A) compared to **buffered baiting** (D)
- No success in disease eradication and spread prevention by acute treatment of **infected area** (B)
- Late success in disease eradication and no spread prevention with **current cumulative strategy** (C)
- Strains of low to intermediate virulence are in the scope of maximum success in spread prevention and disease eradication
- Strains of high virulence are in a scope of potential increase in persistence due to vaccination

### Conclusions

- Disease outcome and virulence has crucial impact on efficacy of oral mass vaccination.
- Strategies currently applied to control CSF in wild boar populations perform suboptimal in virus eradication and spread prevention.
- Preventive strategy, i.e. buffered baiting, is particularly preferable with marker vaccines.
- Vaccination can facilitate disease persistence under high virulence.

#### Acknowledgements

The research leading to these results has received funding from the European Community's Seventh Framework Programme (FP7/2007-2013) under grant agreement n°227003 CP-FP (CSFV\_goDIVA).

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# Neospora caninum antibody detection in serum and milk from Estonian dairy herds



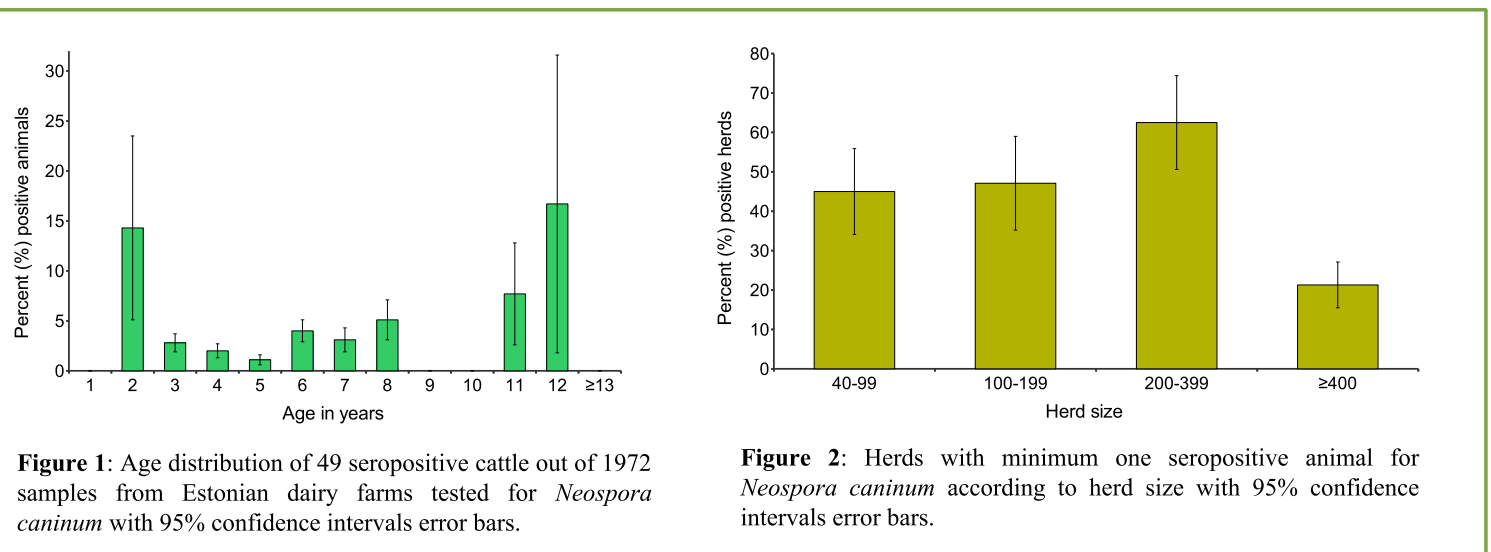
**Brian Lassen, Annely Aleksejev, Kerli Raaperi, Toivo Järvis, Arvo Viltrop, Toomas Orro**  
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## INTRODUCTION

Acknowledgement of *Neospora caninum* (Nc) as an important, but enigmatic, parasitic infection in cattle has steadily increased since its finding in 1984. Increased incidences of abortion and other reproduction problems have been associated with the pathogen worldwide. Infection rates vary greatly between countries and herds [1]. This study gives the first look into larger Estonian dairy herd infection status with Nc.

## MATERIALS AND METHODS

The study consists of: 320 herd bulk milk samples, 1972 individual serum samples of 19-20 cattle between 1-14 years from 100 farms, and additional 119 individual samples from two farms with high milk OD for estimation of maximum in-herd prevalences. Samples were collected from Estonian herds between 2007 and 2008. All samples were randomized and tested for presence of antibodies against Nc using an iscom-ELISA (SVANOVIR® *Neospora*-ab ELISA, SVANOVA). Milk samples were diluted 1:2 and tested as singles, and serum 1:100 and tested as doubles. Cut off value = 20% of the kits positive control (PP). Herd sizes were statistically compared using chi-square analysis.



**Figure 1:** Age distribution of 49 seropositive cattle out of 1972 samples from Estonian dairy farms tested for *Neospora caninum* with 95% confidence intervals error bars.

**Figure 2:** Herds with minimum one seropositive animal for *Neospora caninum* according to herd size with 95% confidence intervals error bars.

## RESULTS AND DISCUSSION

*Neospora caninum* sero-positive animals were found in 2.5% (95% CI: 1.8-3.2) of the animals, while 37.0% (95% CI: 27.4-46.6) of the herds had minimum one seropositive animal. Bulk milk samples tested positive in 16.3% (95% CI: 12.3-20.3) of the herds, being very similar to Latvian reports [2]. Seropositive animals dominate among 1-2 or 12-13 year old animals (Figure 1), which may not contribute milk production as the intermediate age groups which explain the difference between herd prevalences seen with the two methods. Serum samples collected from two herds ( $n=79$  and  $n=40$ ) with the highest bulk milk PP of 53 and 34 respectively resulted in 25 and 35% of the animals being seropositive. The within-herd prevalence of Nc in these farms was high when comparing to Swedish reports [3]. Herds with 400 cows or more animals tended to be less likely to be positive ( $p=0.06$ ), but it is unclear if this is due to management, age distribution or other factors (Figure 2).

## CONCLUSIONS

- 2.5% of cattle were positive for *Neospora caninum* antibodies
- 37% of dairy herds had at least one seropositive cow
- There is a tendency of herds larger than 400 cattle to have less infected animals.
- Within-herd prevalences can reach up to 35%
- 16% of dairy herds tested positive from bulk milk samples

## ACKNOWLEDGEMENTS

Funding for the study was provided by "Cattle health and welfare – aspects of precision livestock farming" (SF0170165s08) and "Measuring of host inflammatory response as a research tool in clinical veterinary science" (8-2/T9001VLVL). Thanks to all veterinarians and farmers who helped collecting the material.

# A multiphase model to estimate lactation milk losses associated with an elevated somatic cell count in early lactation



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<sup>2</sup>School of Veterinary Science, University of Bristol

<sup>3</sup>Pfizer Animal Health Euafme, Paris, France



## Introduction

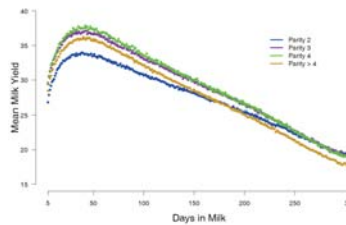
A significant number of mastitis cases occurring in early lactation have been shown to originate from the dry period. While numerous studies have investigated the impact of mastitis on milk production, there exists no estimate of lactation milk losses associated with mastitis in early lactation. Movements across a somatic cell count (SCC) of 200,000 cells/mL between the last milk recording in a lactation and the first milk recording in the subsequent one have been associated with significantly different risks of mastitis and can be used as a proxy for mastitis. The aim of this study was to **model the association between 4 categories of SCC level defined by a threshold of 200,000 cells/mL recorded on the last milk recording of a lactation and the first milk recording in the following lactation on the shape of the lactation curve.**

## Study

### Data

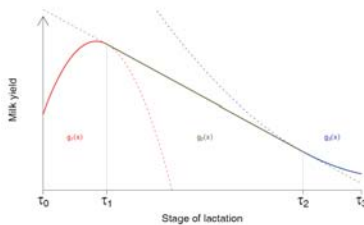
- Monthly collected milk recording data
- England and Wales
- January 2004 to December 2006
- 8 million individual cow recordings**
- 1 million lactations**
- 0.5 million cows**
- 2,128 herds**

### Mean milk production per day in milk



- Milk production exhibits multiple phases:
  - Ascending phase: calving to ~ 50 days
  - Descending linear phase: 50 to 250 days
  - Non linear after 250 days

## A multiphase model for lactation curves



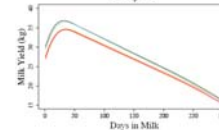
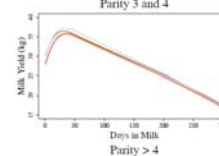
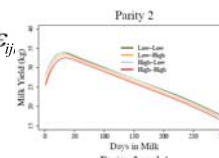
Lactation curves can be modelled using piecewise functions

- Cubic function for the ascending phase
- Linear decrease after the peak
- Quadratic function for the end of lactation

$$f(x) = \begin{cases} g_1(x) = \alpha_0 + \alpha_1x + \alpha_2x^2 + \alpha_3x^3 \\ g_2(x) = \delta_0 + \delta_1x \\ g_3(x) = \gamma_0 + \gamma_1x + \gamma_2x^2 \end{cases}$$

This is equivalent to fitting the following multilevel model:

- $$Y_{ijk} = \beta_0 + \beta_1x_{ijk} + I_1(\beta_2x_{ijk}^2 + \beta_3x_{ijk}^3) + I_3\beta_4(\tau_2^2 - 2\tau_2x_{ijk} + x_{ijk}^2) + \varepsilon_{ij}$$
- $Y$  : milk yield
  - $x$  : days in milk
  - $i$  : recording level
  - $j$  : lactation level
  - $k$  : herd level
  - $\tau_2$  : second change point
  - $\varepsilon$  : error term. The error was split between herd, lactation and residual levels
  - $I_1$  : indicator variable 1 in interval 1 ; 0 otherwise
  - $I_3$  : indicator variable 1 in interval 3 ; 0 otherwise
  - Days in milk were centred on  $\tau_1 = 60$  days in milk
  - Models were fitted on a random sample of 19,799 lactations from 1,000 herds



- Curves fitted for 4 SCC categories and 3 parity categories
  - Low or High: below or above 200,000 cells/mL
  - On the Last milk recording of previous lactation – First milk recording of the lactation modelled (e.g. Low-Low)
- Difference in milk production between categories predicted by the model:

	Parity 2	Parity 3 and 4	Parity > 4
Low-Low (Ref)	8032	8477	8380
Low-High	-198	-177	-400
High-Low	-23	120	-86
High-High	-366	-162	-493

## Main findings

A multilevel multiphase model was found to fit lactation curves accurately when looking at the mean of multilevel residuals per day in milk (not shown – submitted for publication and available on request). The intercept ( $\beta_0$ ) of this model corresponds to the milk production at 60 days in milk and the coefficient for days in milk ( $\beta_1$ ) to the persistency. The model predicted a milk production between 162 and 493 kg lower in cows with a SCC > 200,000 cells/mL during the first month after calving as compared to cows with a SCC < 200,000 cells/mL on both the last milk recording of the previous lactation and the first milk recording in the current lactation.

# RANDOM FORESTS TO EVALUATE ANTIMICROBIAL MULTIRESTANCE IN SALMONELLA TYPHIMURIUM

Marzia Mancin, Marco Ruffa, Lisa Barco, Antonia Anna Lettini, Antonia Ricci  
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## INTRODUCTION

Antimicrobial resistance in zoonotic bacteria is a problem of increasing concern worldwide and multiresistance is a critical aspect of such phenomenon, since it can lead to human infections untreatable with available drugs. In order to counteract the diffusion of resistance, scientists have to provide policy makers with relevant data on factors contributing to its emergence and spreading, and statistical tools like Random Forests (RF) are used to guarantee the significance of results and to facilitate their interpretation.

## MATERIALS AND METHODS

- > A selection of 556 *S. Typhimurium* strains isolated in Italy from different types of sample (animals, foodstuffs and environment) during the period 2004-2009 was submitted to antimicrobial resistance analysis by disk diffusion;
- > A panel of 15 antibiotics was used to evaluate the antimicrobial multiresistance (AMR), defined as resistance to more than 3 antimicrobials;
- > RF was used to verify which variables among years, districts, material and place of sampling, species, type of sample and phagetype are more important to explain the AMR in terms of mean decrease accuracy (MDA) and mean decrease of Gini (MDG)
- > The AMR was considered as:
  - binomial variable: 0=no multiresistant, 1=resistant to more than 3 antimicrobials; i.e. 2 way classification;
  - multinomial variable: 0=no multiresistant, 2=resistant to more than 3 antimicrobials, 1=otherwise; i.e. 3 way classification; resistant to K=0, 1...6 antimicrobial; i.e. K way classification.

## RESULTS

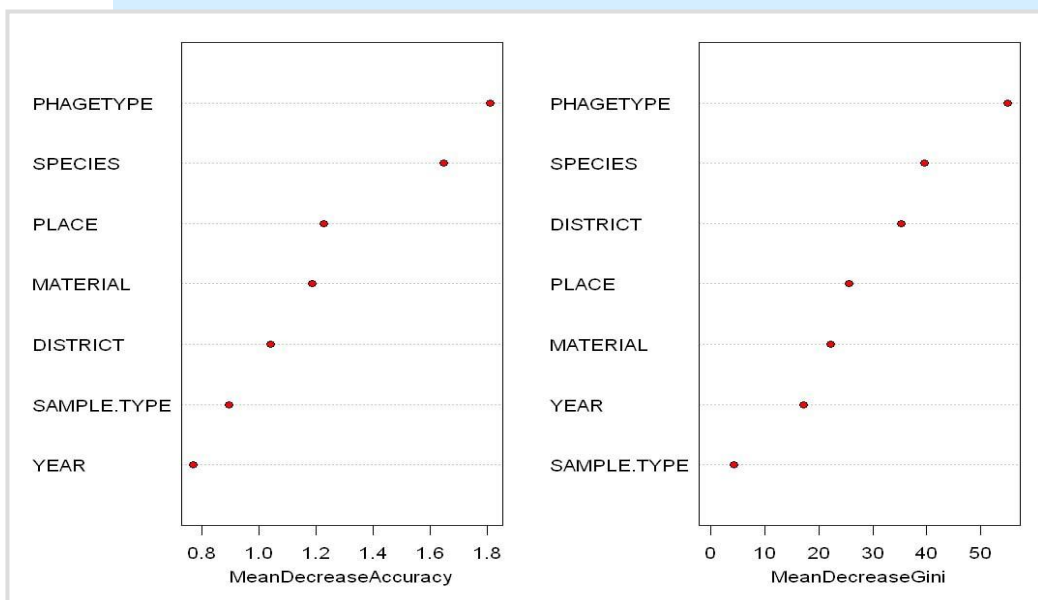


Figure a: Classification of covariates for the AMR in accordance with MDA and MDG.

RF output shows that phagetype and species are two covariates that explain most of the AMR, whereas year and type of sample are not important (Fig. a). To evaluate the accuracy of classification method it is possible to design Multi-dimensional Scaling Plot to visualize the dissimilarity data in a two dimensional space and the convergence of error that shows the errata classification of model per category (Fig. b). In the case of binomial AMR it is also possible to design the ROC curve (Fig. c). The increase of AMR classification way produces a rise in average rate of error classification (Tab. 1).

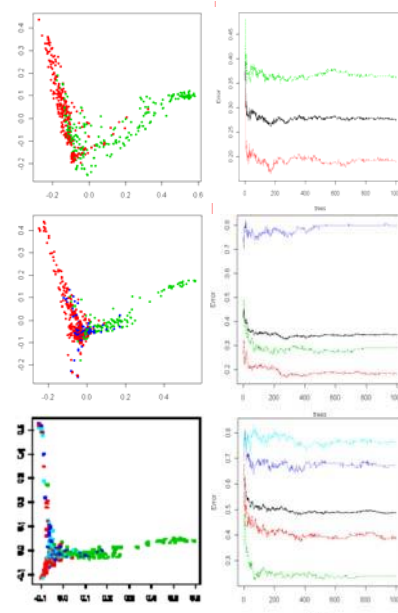


Figure b: Multi-dimensional Scaling Plot and Convergence of error per category in 2-way, 3-way and 4-way analysis.

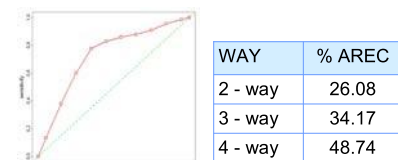


Figure c: 2 - way : ROC curve. Table 1: Average rate of Error Classification of Random Forest.

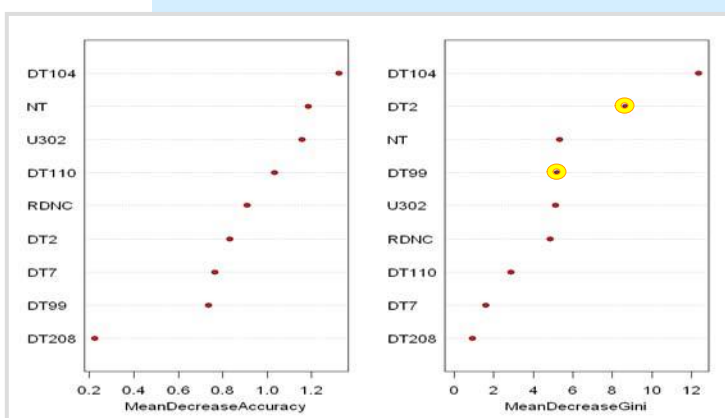


Figure d: Classification of covariates for the AMR in accordance with MDA and MDG.



Figure e: Classification tree of phagetypes.

Phag.	LDA	LR	p-value
DT104	2.04	1.82	0.000
NT	1.04	0.82	0.000
U302	1.92	1.68	0.001
DT110	2.47	2.53	0.017
DT7	1.70	1.43	0.037
RDNC	-0.35	-0.29	NS
DT2	-2.13	-14.86	NS
DT99	-2.13	-14.87	NS
DT208	-0.74	-0.64	NS

Table 2: Coefficients of LDA and LR regression.

RF applied to different phagetypes shows that DT104, NT, U302, DT110 (multiresistant) are the most significant phagetypes for the prediction accuracy of model (MDA) of AMR; MDG indicates that, besides these variables, also DT2 and DT99 (not multiresistant) are important for the splitting of dependent variable (Fig. d). RF is consistent with the results of the classification tree (Fig. e), of the Linear Discriminant Analysis (LDA) and of the Logistic Regression (LR) (Tab. 2) or Multinomial Regression (MR) (2 or more way respectively). The classification error for these different methods is in this case very similar.

## CONCLUSIONS

RF yield manageable results, allow to analyze high amounts of variables and to assure non parametric robust results. RF represents a valid alternative to other methods of multivariate analysis such as LDA and MR.

# Why limit yourself to one logistic regression model?

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## Introduction

Choice of a regression model usually relies upon use of a forward selection or backward elimination process. This may lead to equivalent models being overlooked particularly in observational studies.

A preferable approach is to fit all possible models and evaluate them. This can be a useful approach where sample size is limited although care must be taken in selection of the terms to be incorporated in the procedure.

Various programs directly offer this procedure (e.g. R's `bestglm`; Genstat's RSEARCH) but it can be carried out in other programs (Stata, SAS, SPSS).

The following example illustrates a study where more than one model is relevant (otherwise information would be lost).

## Example

### Methods outline

A regional case-control study on the herd risk factors for the disclosure of tuberculin skin test reactors was carried out on a random sample of cattle herds where badger activity had been surveyed (cases = 77 herds ; controls = 108 herds). Main risk factors are listed in the table below.

Cases = Herds that did have one or more TB test reactors during a 3 year period.

Controls = Herds that did not have any TB test reactor during a 3 year period.

GenStat® used for statistical analyses:  
Logistic regression ( $P \leq 0.15$  cut off for including in multivariate model) and RSEARCH procedure used to select best fit models.

The eight best subsets for inclusion of a different number of terms (2-5) were examined

## Results/Discussion

Three models were deemed to be biologically plausible (see table).

### Best subsets with 3 terms

Akaike Information Criterion	Schwarz Information Criterion	DF	Previous TB history	Badger activity	Herd Size	Market to Farm Movements	Cattle Tracing
212.02	231.34	6	0.001	0.032	0.001	-	-
219.89	232.77	4	0.000	0.006	-	0.016	-
219.92	232.80	4	0.000	0.002	-	-	0.017

The three models consistently indicated that a previous history of TB in a herd and the observation of badger activity on the farm were risk factors for the presence of TB in a herd. The third term was dependent upon which model was used, with all three considered as valid risk factors (increased herd size, cattle movements onto the farm from markets and epidemiological tracing of cattle).

## Conclusions

- This approach allows you to look at all possible model permutations and highlights other logistic regression models that are worthy of consideration.
- In the example, two valid risk factors would be ignored if more traditional methods were used.

# Monitoring of antibiotic use in livestock in Germany: Results of a feasibility study

Merle R<sup>(1)</sup>, Hajek P<sup>(2)</sup>, Käsbohrer A<sup>(3)</sup> and Kreienbrock L<sup>(1)</sup>

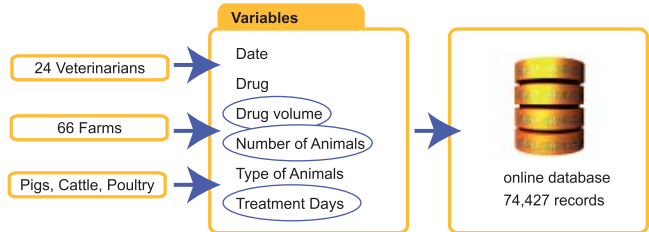
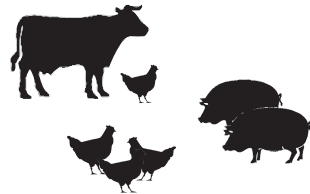
<sup>1</sup> WHO Collaborating Centre for Research and Training in Veterinary Public Health, Department of Biometry, Epidemiology and Information Processing, University of Veterinary Medicine, Hannover, Germany. <sup>2</sup> Department of Pharmacology, Pharmacy and Toxicology, Veterinary Faculty of Universität Leipzig, Germany. <sup>3</sup> Federal Institute for Risk Assessment, Berlin, Germany.

## Introduction

The amount of applied antibiotics may be linked to the development of bacterial resistances. The drug's dose and the application period are regarded as the most important factors affecting the spread of bacterial resistances.

A monitoring system concept to evaluate data of antibiotic use in livestock by a bottom-up approach was tested in a feasibility study.

## Material and methods



Substance(s)

Substance mass

$$\# \text{ Applications} = \# \text{ Animals} * \text{Treatment days} * \# \text{ Substances}$$

$$\text{Treatment frequency}_{\text{Animal \& year}} = \# \text{ Applications} / \text{Population size}^1$$

<sup>1</sup>) Population size: e.g. all animals per farm or per district

## Results

### Antibiotic use

Tetracyclines were the most frequently used substances. But regarding the number of applications, the percentage of tetracyclines was smaller, while the percentage of macrolides and of the sulfonamides/trimethoprim group was higher (figures 1, 2). For monitoring purposes, the number of applications is regarded more important than the consumption in kg.

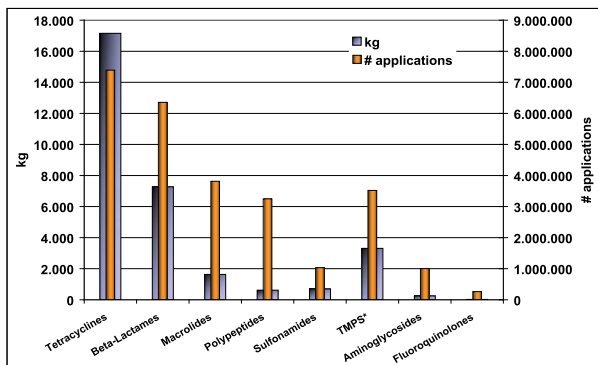


Figure 1: Antibiotic use in pigs \*TMPS: Trimethoprim & Sulfonamides

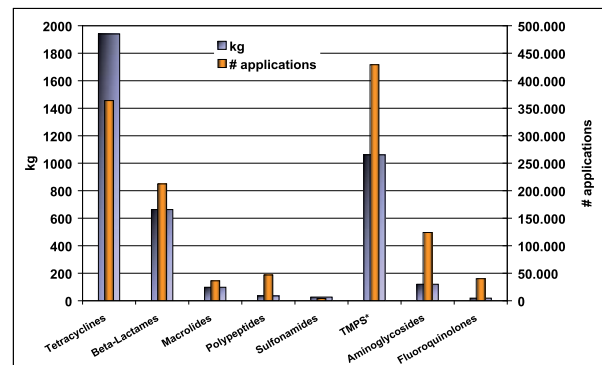


Figure 2: Antibiotic use in cattle

### Treatment frequency

- ★ Data from farms: easy calculation.
- ★ Data from veterinarians: Treatment frequency could not be calculated, because the population size (i.e. all animals attended by the veterinarian) was not known.
- ★ Pigs are treated more often than cattle (table 1).

The treatment frequency is suitable for monitoring systems, because it is related to the population and can be compared between different regions or from year to year.

Table 1: Treatment frequency (Geometric mean of farms per animal & year)

Fattening Pigs*	8.9
Dairy cattle	2.5
Calves	2.3

\* per fattening pig (30 kg until slaughter)

## Feasibility

Although several information gaps were identified, e.g. the number of treated animals per veterinarian, the proposed system is able to function as a monitoring system for antibiotic use in Germany.

A pilot study will take place in 2011 including eight German districts in order to gain representative monitoring data.

# Estimating Young Stock Rearing Total Costs using Stochastic Simulation

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## Introduction

- Young stock rearing is often being overlooked due to unawareness of Dutch farmers to the total costs.
- Previous studies on the total costs estimation did not consider continuous and variation in growth and uncertainty in disease incidence.
- Several costs component (feed, healthcare, breeding) are interrelated with variation in growth and uncertainty in disease incidence.

## Objective

- Estimating the distribution of total costs of young stock rearing in The Netherlands using stochastic simulation.
  - A bio-economic model is developed.
  - Variation in growth and uncertainty in occurrence of diseases is included.

## The Bio-economic model

- Add-in @Risk in Microsoft Excel.
- Stochastic simulation with 20,000 replications.
- No disease transmission or herd dynamics.
- 56 stages to reflect age.
  - Simulating at calf level from 2 weeks until first calving.
- States to reflect biological changes in young stock life.
  - Bodyweight.
  - Health status: Healthy, diseased, death.
    - Two diseases included: Calf Scours (CS) and Bovine respiratory diseases (BRD).
  - Breeding status: Pre-pubertal, cyclic, pregnant.
- Transition between states within stages.
  - Growth curve unique for every simulated calf using two phase growth function (Koenen and Groen, 1996).
  - Health status is determined by using transition matrices (Table 1) built at every stage (56 transition matrices).
  - Disease effect (growth rate reduction) is based on age and has only short term effect.
- Economic components.
  - Based on literature and expert knowledge.

Table 1: Example of a transition matrix to simulate health status at 2 weeks of age (stage 2).

		Week 2 (Stage 2)			
Week 1 (Stage 1)	States	Healthy	CS	BRD	Mortality
	Healthy	0.9536	0.044	0.0024	0
	CS	0.70	0	0	0.38
	BRD	0.88	0	0	0.12
	Mortality	0	0	0	1

## Results

- The total costs of rearing a young stock is € 1540.
- The total costs of young stock rearing is approximately 13% of the cost price of milk.
- Average first calving age is 25 months with average weight of 542kg.
- The total costs of rearing is €78 higher when a heifer was diseased once compared to a non-infected heifer.
- Diseases do not affect first calving age and weight.
- Growth rate affect first calving age, weight and total costs of rearing.

Table 2 : The costs of rearing a young stock that successfully reached first calving age (5% and 95% percentiles given in brackets).

Costs component	Average costs (€)
Calf price	55
Prevention	26
Farmer's treatment	2 (0; 36)
Veterinary treatment	5 (0; 0) <sup>1</sup>
Feed	697 (657; 778)
Labor	499 (475; 536)
Breeding	40 (27; 81)
Barn	180 (176; 198)
Mortality and culling	34
Total	1540 (1423; 1689)

<sup>1</sup> The 1% to 99% percentiles is €0 to €186

## Average total rearing costs change

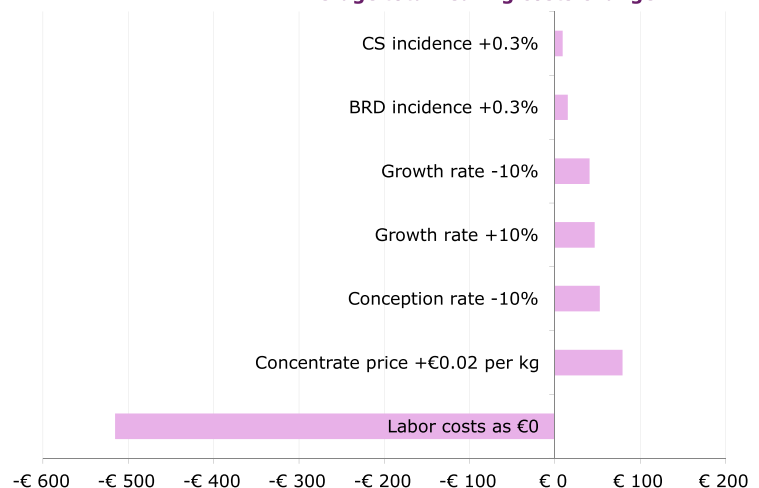


Figure 1: Sensitivity of average total costs of young stock rearing for disease incidence, growth rate, conception rate, feed costs and labor costs.

## Conclusions

- The total costs of rearing a young stock is € 1540.
- The total costs of young stock rearing is approximately 13% of the cost price of milk.
- Healthcare and mortality costs are minimal costs in average total rearing costs.
- Optimal growth is important to reduce total rearing costs.
- The model is able to calculate total costs and give insights in young stock rearing total costs distribution.
- Farmers should be aware of the total costs and should make management changes based on important findings of this study.

## Reference

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# More Evidence of Interspecies Transmission of *Mycobacterium avium* subsp. *paratuberculosis* Between Livestock and Wildlife: The Hare



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## Objectives

The aim of this study was to evaluate the potential association between the prevalence of *Map* infection in cattle and the prevalence of *Map* infection in hares, in southern Chile.

## Material and Methods

• A case-control study was performed in southern Chile. A highly infected dairy herd (A) with historical information about disease status (10 cases/year with clinical cases and infection confirmed by fecal culture) was used as case. A second herd (B) without history of clinical disease, serologically negative at least at two consecutive testing one year apart and located more than 300 km apart from the case herd, was used as control herd.

• 50 hares from herd (A) and 42 from herd (B) were captured and processed for *Map* detection from ileum (IL), mesenteric lymph nodes (MLN) and fecal samples (FS).

• Hares were hunted in 2 rounds in each herd by professional hunters, during June and July 2009 and the location of hares at the moment of capture was geo-referenced.

• Cattle pooled fecal samples from 5 animals were obtained from a representative group of the older lactating cows (150).

• Soon after hunting, hares were quickly transported to the necropsy unit at the Faculty of Veterinary Sciences, where representative tissue samples were aseptically collected. Samples were decontaminated and processed following manufacturer's protocols for cultivation in an automated liquid culture system (BACTEC MGIT 960). After incubation at 37 C for 49 days, DNA was extracted from all positively signaling tubes, and real-time PCR based on the IS900 and MAP02 genes was performed, to confirm MAP. Age of hares was estimated based on dried weight of optical lens.

The association between characteristics of hares (sex, age), location (Farm A vs. B) and round of capture (first, second) and disease status, were assessed by logistic regression (LR) adjusted by the estimations of diagnostic performance.

## Conclusions

We found evidence for interspecies transmission, but the proved association is not necessarily causal.

Absence of histopathological lesions might indicate absence of disease.

The results improve the epidemiological knowledge on paratuberculosis regarding interspecies transmission and the concept of livestock as a source of infection for wildlife.

## Results

• In herd A, 45 individuals had one or more type of sample signaling culture positive, whereas in herd B, 17 hares showed at least one positive culture result ( $P < 0.05$ ).

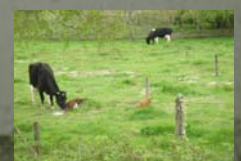
• Regarding pooled samples of bovine feces, herd A showed 20 positive samples out of 30, compared to 12 out of 30 for cows from herd B. Cattle herd and hare prevalences of infection were statistically significant ( $P < 0.05$ ).

• MAP infection was detected in MLN of hares one to four months old. Tissue samples positive for MAP in hares with FS positive culture varied as age increases, however, oldest categories (8-12 months and >12 months) seems to present more proportion of infected tissues.

• Also, more hares captured from herd A were infected and in these hares at least one tissue was MAP+, in comparison with those from herd B. In addition, infected individuals from all ages categories were detected in herd A, whereas only old animals in herd B.

• After checking for interactions and potential confounders the final LR model contained 3 variables, however, only the variable "farm where hare was cropped" was statistically significant.

The work was supported by FONDECYT Project # 1085069



# Risk factors for Infectious Salmon Anemia (ISA) outbreaks and virus transmission in farmed salmon from Southern Chile



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## Objectives

The aims of the study were:

- A) to identify host, environmental and management risk factors associated with ISAv outbreaks for a year class of Atlantic salmon under the current conditions of the Chilean production system;
- B) to assess space-time clustering of outbreaks.

## Conclusions

- More outbreaks occur late in the 1<sup>st</sup> year of the marine-phase.
- More net pen-level variables than site-level variables were statistically associated with risk of outbreak.
- There was space-time heterogeneity that needs to be considered

## Material and Methods

- The retrospective study with all smolts stocked from a company (01/ 2008 until reaching commercial weight or by stamping out due to ISA.
  - **A case was defined** as a net-pen that presented fishes with clinical signs (or lesions consistent with ISA), a disease-induced mortality rate of at least of 0.01% and at least one fish testing positive to reverse transcription-polymerase chain reaction test for ISAv.
  - **A suspected net-pen** was one that fulfills at least one of the 3 criteria above or if it had received live fishes from another suspected or confirmed case site or net-pen.
  - A personally administrated questionnaire was applied to each veterinarian responsible for the site and it contained several questions related to site-level and net-pen-level, for production conditions, environmental, health and management practices, etc.
  - The incidence rate of outbreaks was estimated as the number of new cases over the number of net-pens-month-at-risk, on a monthly basis.
  - A Cox proportional hazard model containing random and fixed effects was used to evaluate potential risk factors.
- Finally a space-time retrospective analysis (using cases only) was performed using SATScan.

## Results

All sites presented outbreaks and 139/243 (57.2%) net-pens were defined as cases .

Median time from stocking time to outbreak was 251 days, IQR= 142.

The final model contained 9 main effects and 3 interactions (but only one statistically significant), those that increased the risk of outbreak ( $P < 0.05$ ) were:

- total no. stressing events (HR 1.31),
- total no. outbreaks of vibriosis and Salmon Rickettsial Syndrome, previous to ISA outbreak (HR 2.06),
- no. sea lice treatment-baths (HR 3.37),
- no. husbandry practices applied on the net-pen (HR 1.89).

Variables associated with a decreased risk of an ISA outbreak ( $P < 0.05$ ) were:

- average stocking weight (net-pen) (gr) (HR 0.99),
- proximity (mt) to a net-pen with an outbreak (HR 0.98),
- site not sharing wharfs (HR 0.005),
- plant B where fish is processed (HR 0.19);
- and surveillance district in which the site is located

Interactions

- Total No. stressing events\* No. sea lice treatment-baths (HR=0.96)

The space-time analysis detected a cluster composed by 2 sites during 09/2008 to 12/2008.



# Novel method accurately predicts the number of cats owned by households in Wales



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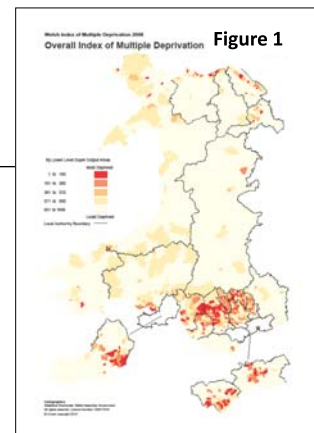
**SUMMARY:** Data from a large (n=4549) survey were used to derive estimates for the number of cats owned by households in Wales in 2009-2010 using four different methods, including a novel method based on the 2008 Welsh Index of Multiple Deprivation (WIMD). It was possible to assess the accuracy of three methods, by comparing actual and predicted numbers of cats owned in a random sample of 10% of the dataset, using models constructed from the remaining 90% of the dataset. All three methods predicted the number of cats owned with a high level of accuracy. These results demonstrate that the WIMD can be used to predict cat ownership, which will be of particular relevance to those wishing to use postcode data to predict cat ownership in different areas of Wales.

## BACKGROUND

- Different methods have been used to estimate the number of cats and dogs the UK (Westgarth *et al.*, 2007; Downes *et al.*, 2009; Murray *et al.*, 2010); however in the absence of a gold standard it is difficult to assess the accuracy of these methods.
- Previously reported methods use predictor variables that are available from census records, in order that predictions of cat and dog ownership can be made. This study investigated a novel method of predicting cat and dog ownership, by using the 2008 Welsh Index of Multiple Deprivation (WIMD 2008).
- The WIMD 2008 is a relative measure of deprivation at the small area level in Wales (Figure 1). WIMD is constructed from eight different factors related to deprivation with the following weightings (Welsh Assembly Government, 2010):

Income (23.5%)	Employment (23.5%)	
Education, skills and training (14%)	Health (14%)	Geographical access to services (10%)
Community safety (5%)	Housing (5%)	Physical environment (5%)

- The Welsh Assembly Government (WAG) conducted a face-to-face survey of a sample of 4559 Welsh households during 2009-2010.



## AIMS OF THE STUDY

1. To estimate the number of cats owned by households in Wales using four different methods
2. To compare the accuracy of these four methods.

### Method 1: Simple method of extrapolation

The mean number of cats owned within the study sample was multiplied by the total number of Welsh households (n=1,338,811) to produce an estimate of the size of the pet cat population in Wales.

### Method 2: Model-based approach. Using number of people in household as a predictor.

SPSS was used to fit a linear regression model to the data. Five categories were used (1, 2, 3, 4, 5 or more people in the household). For each category estimates of the average number of cats and the standard errors were constructed (Table 1). Estimates were then combined into a weighted sum based on the number of Welsh households in each of the 5 household sized categories to estimate the total population of pet cats in Wales.

### Method 3: Model-based approach. Using Welsh Index of Multiple Deprivation (WIMD) as a predictor.

The same approach was used, as described for Model 2. However, instead of using the number of people in the household, two categories of WIMD 2008 were used in the model (Table 2); households in the 40% most deprived areas of Wales and households in the 60% least deprived areas of Wales. These two categories resulted in the best model fit.

**Method 4: WAG predictions.** The figure for pet cat population in Wales is based on the actual number of cats per household in the study sample, grossed to the total number of households in Wales, adjusted for sampling weights.

**Table 1:** Based on complete dataset (n=4549)

No. of people in household	Coefficient	Standard Error
1	0.174	0.017
2	0.210	0.015
3	0.316	0.027
4	0.352	0.031
5 or more	0.402	0.045

**Table 2:** Based on complete dataset (n=4549)

WIMD 2008 of household	Coefficient	Standard Error
40% most deprived	0.207	0.015
60% least deprived	0.253	0.013

## Table 3: RESULTS

	Estimated number (95% CI) of pet cats in Wales Complete dataset (n=4549)	Estimated number (95% CI) of pet cats in Wales 90% of data randomly selected (n=4094)	Estimated number (95% CI) of cats in 10% of data (n=455) excluded from original dataset 107 cats owned by households
Method 1 (Mean no. of cats/household)	314,391 (288,803-339,979)	314,264 (287,178-341,351)	107 (98-116)
Method 2 (No. of people in household)	337,346 (311,113-363,579)	336,426 (308,589-364,264)	108 (99-117)
Method 3 (WIMD 2008)	315,833 (290,226-341,440)	315,760 (288,654-342,866)	107 (98-116)
Method 4 (WAG predictions)	344,564 (319,140-369,988)	n/a	n/a

## DISCUSSION:

Methods 1-3 were able to predict the number of owned cats with a high level of accuracy, as demonstrated in Table 3. The use of WIMD 2008 to predict pet ownership is a new finding which has potential uses in many areas as it is not dependent upon data collected from individual households. Potential areas of use include: identifying areas which are more likely to own higher numbers of cats which will be of interest to managers of vet practices, pet shops and feline welfare charities. Further use of the data to include density of the owned feline population may also be useful in predicting areas where the risk of feline infectious disease transmission is greatest.

**ACKNOWLEDGEMENTS:** Cats Protection for funding Jane Murray's post. The Welsh Assembly Government for providing the data for analysis. Questionnaire respondents are thanked for participating in the study.

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# Elimination from elite endurance rides in nine countries in 2008 – a preliminary study

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## Introduction

Endurance is the fastest growing Fédération Équestre Internationale (FEI) discipline.

The high elimination rate from endurance rides has been a regular matter for discussion in professional and public forums.

In recent years, elimination rates seem to have increased.

## Objectives

To document elimination rates and to explore potential risk factors for elimination due to lameness or metabolic reasons in 9 countries representing all 5 continents

## Hypothesis

Weather conditions and terrain considerably vary between countries; therefore we hypothesised that country would have a significant effect on elimination rates for both lameness and metabolic reasons.

## Materials and Methods

Data for 4326 started horses in endurance rides of  $\geq 100$  km held in 2008 in Australia, France, Italy, South Africa, Spain, United Arab Emirates (UAE), United Kingdom (UK), Uruguay and United States of America (USA) were collected from the FEI website ([www.fei.org](http://www.fei.org)).

For each class, the country, the distance of the ride, whether the class was restricted to young riders, the number of started horses and the winning speed were recorded. Horses that retired or were eliminated for lameness, metabolic or other reasons were recorded.

Univariable and multivariable logistic regression models were used to assess the effect of country, number of horses in the class, ride distance and young rider class on two outcomes (elimination for lameness and metabolic reasons).

**Table 1.**

The number of horses that started and completed the ride or were retired or eliminated at Fédération Équestre Internationale endurance rides of  $\geq 100$  km in 2008 in nine countries. The percentages indicate the proportion of all started horses. N = number

Country	Started N	Completed N (%)	Retired N (%)	Eliminated N (%)			
				All	Lameness	Metabolic	Other
Australia	155	99 (63.9)	1 (0.7)	55 (33.7)	46 (29.7)	5 (3.2)	3 (1.9)
France	1029	565 (54.9)	85 (8.3)	380 (36.9)	297 (28.9)	63 (6.1)	20 (1.9)
Italy	291	148 (50.9)	21 (7.2)	122 (41.9)	81 (27.8)	22 (7.6)	19 (6.5)
UK	287	124 (43.2)	20 (7.0)	143 (49.8)	113 (39.4)	17 (5.9)	13 (4.5)
South Africa	243	140 (57.6)	17 (7.0)	86 (35.4)	61 (25.1)	16 (6.6)	9 (3.7)
Spain	408	180 (44.1)	48 (11.8)	180 (44.1)	110 (27.0)	51 (12.5)	18 (4.1)
UAE	1497	510 (34.1)	140 (9.4)	847 (56.6)	544 (36.3)	255 (17.0)	45 (3.0)
Uruguay	240	111 (46.3)	7 (2.9)	122 (50.8)	83 (34.6)	29 (12.1)	10 (4.1)
USA	176	112 (63.6)	10 (5.7)	54 (30.7)	45 (25.6)	9 (5.1)	0 (0)

## Results

Data for 157 classes at 91 events were analysed. The UAE had the highest number of entries (1497), followed by France (1029) and Spain (408).

Of 4326 started horses, 46.0% finished the ride (Table 1).

Lameness was the most common cause of elimination in all countries followed by elimination for metabolic reasons (69.2% and 23.5% of all eliminations and 31.8% and 10.8% of all started horses, respectively).

Eight % of horses were retired by the rider, having passed the veterinary examination.

In multivariable analysis, the risk of **elimination for lameness**

- was associated with the country in which the ride was held; horses competing in rides held in the UK (odds ratio [OR] =2.11, P<0.001), UAE (OR=1.72, P=0.007) and Uruguay (OR=1.69, P=0.013) had the highest risk of elimination for lameness.

- the risk significantly increased (OR=1.60) for horses competing in rides with  $\geq 80$  entries.

The risk of **elimination for metabolic reasons**

- was also significantly associated with the country in which the ride was held; the highest risks of elimination for metabolic reasons were recorded in the UAE (OR=7.28, P<0.001), Spain (OR=5.61, P<0.001), Uruguay (OR=5.26, P<0.001) and Italy (OR=2.94, P=0.035).

- the risk significantly increased (OR=2.17) for horses competing in rides with  $\geq 100$  entries.

## Discussion

To date, this is the largest study describing completion rates and risk factors for eliminations due to lameness and metabolic reasons from endurance rides in different countries.

The country of the ride had a significant effect on the risk of elimination due to both lameness and metabolic reasons. The number of entries in the ride was also significantly associated with the risk of elimination due to either lameness or metabolic reasons.

This may be attributable to a number of factors, including terrain, weather, training, riding (e.g. speed and fitness), and quality of veterinary control at the ride; all of which require further detailed investigation.

This study was carried out as a pilot study for a prospective large-scale, world-wide study that aims to investigate risk factors for lameness and specific orthopaedic injuries resulting in elimination from elite endurance rides. The overall aim is to improve the welfare of endurance horses, and to help to base the FEI ride regulations on evidence-based data.

## Conclusions

- Elimination rates varied between countries, with lameness being the most common reason for elimination globally.
- The country and the number of started horses were risk factors for elimination due to lameness and for elimination due to metabolic disorders.
- A prospective study is needed to assess the effects of environmental conditions, individual horse speed and other variables on the risk of specific causes of elimination.





# Milk Yield and *Salmonella* in Dairy Cattle Herds



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## Background and Objective

*Salmonella* infection in cattle has long been reported to affect milk yield. However, no studies to date have quantified milk yield loss over an extended period of time or estimated how long it takes before yield is back to pre-infection levels.

**Objective:** To quantify milk yield losses associated with *Salmonella* in dairy cattle herds

## Materials and methods

**Study population:** 68 Danish Holstein herds, >40 cows in total included, in total 11.959 cows

**Case herds (n=28)**

Herds active between 2005-2009 with a minimum of 1 year of low bulk-tank milk *Salmonella* antibody levels (<10 ODC%) followed by increase to high *Salmonella* antibody levels (≥70 ODC%).  
 Estimated infection date = date of high bulk-tank milk value - 61 days

**Control herds (n=40)**

Low bulk tank milk antibody levels (<10 ODC%) 2005-2009  
 Artificial infection date weighted by year and month to match case herds  
 Control herds were used to evaluate whether the effects in the case herds could be reproduced in herds without *Salmonella* infection

### Statistical analysis:

Daily milk yield (kg ECM) modelled in multilevel hierarchical model with 3 levels: yield, cow and herd with repeated measurements for yield recordings. Parity 1, 2, and 3+ modelled separately

Time from estimated herd infection included in 3-month intervals (T), where T<sub>0</sub> was 0-3 months after infection, T<sub>1</sub> was 4-6 months after and T<sub>-1</sub> was 3-0 months before herd infection. Milk yield recordings from 12 months before to 18 months after estimated herd infection (T<sub>-4</sub> – T<sub>5</sub>) were used.

Year, season for milk recording, DIM, Log(SCC) was included as confounders at yield level and average herd size in the study period at herd level  
 Wilmink's function ( $e^{-0.05^{DIM}}$ ) was also included

## Results and Conclusions

### Results:

Reduced yield in parity 1 and 3+ 6-15 months (T<sub>2</sub>-T<sub>4</sub>) after herd infection date (Fig. 1). Mean loss for parity 1 cows was 1.4 kg ECM/cow/day (95% CI:0.5-2.3) and 3.0 kg ECM/cow/day (95% CI: 1.3-4.8) for 3+ parity cows, when compared to cows in same parity before herd infection. Only minor differences before and after the infection date was seen in parity 2 cows

For a herd with 100 "year-cows" with 36, 32 and 32% cows in parity 1, 2 and 3+ respectively, these results indicate a mean reduced yield of 40,000kg ECM (95% CI: 8,000-153,000) in the 18 months after herd infection date

Year, season, DIM and Log(SCC) were significantly associated with daily milk yield, but herd size was not. There was no effect in simulated infection date in control herds

### Conclusion:

Introduction of *Salmonella* to the herd is associated with large reduction in milk yield for cows in parity 1 and 3+. It takes on average about 15 months before yield is back to pre-infection levels

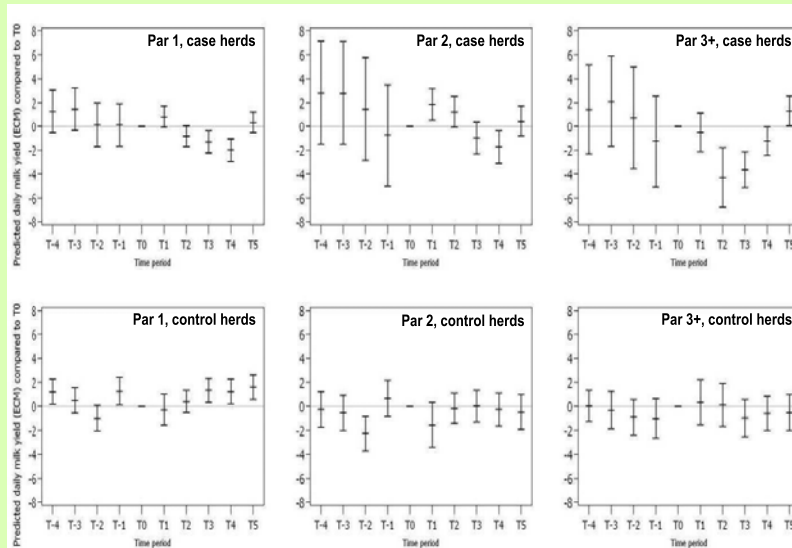


Fig. 1 Predicted mean daily milk yield (Kg ECM) compared to T<sub>0</sub> for case and control herds





## Background

Hepatitis E is an emerging zoonotic disease distributed worldwide. On the basis of clinical and epidemiological observations HEV was initially associated only with acute hepatitis in humans. The disease is self-limiting in the majority of patients. However, high morbidity and mortality rates have been described in pregnant women. Besides in humans the causative agent Hepatitis E virus (HEV) is found in domestic animals such as swine as well as in the wildlife species such as wild boar, deer, rabbits and rats. So far no clinical disease has been associated with HEV in animals.

HEV is a non-enveloped virus with a single stranded positive sense RNA genome of approximately 7.2 kb. The virus belongs to the family *Hepeviridae* which has been subdivided into four genotypes named I-IV according to the sequence of discovery. Members of certain genotypes are described to differ in geographic distribution, clinical and zoonotic aspects. The epidemiological relevance of the assortment to genotypes and subtypes is not completely clear especially since different regions of the HEV genome are analyzed. We presume the use of complete genomic sequences provides more information and will make the analysis more reliable.

The objective of this study was to perform phylogenetic analyzes based on complete HEV genomic sequences in order to evaluate the current taxonomy of HEV at genotype and subtype level.

## Materials and Methods

HEV complete genomic sequences available from GENbank were obtained for selected isolates of human and animal origin.

Sequences were arranged in an alignment of 125 complete HEV sequences. Phylogenetic analyses were performed using the HUSAR package (DKFZ Heidelberg). Phylogenetic distances were calculated (Kimura-2 parameter method) and trees generated based on neighbor-joining and maximum-likelihood methods. Bootstrap analysis with 1000 replicates were included to demonstrate statistical significance. Phylogenetic trees were plotted using ClusTree and TreeView 1.6.6 Software.

## Results

According to our analyses HEV isolates cluster in at least six groups including the four established genotypes (GT I - IV) and a new group containing virus isolates from rabbits ("GT V"). Surprisingly our data show that GT III forms two distinct clusters which were provisionally termed GT IIIa and GT IIIb. The use of complete genomic sequences improved the reliability of the analyses which was supported by high bootstrap values. The phylogenetic trees calculated by the neighbor-joining or the maximum-likelihood method showed an almost identical topology (Fig. 1). Virus isolates obtained more recently significantly contributed to the separation of GT III into two branches.

The maximum nucleotide distances observed within genotypes were 13.16% (GT I), 20.02% (GT IV), 17.91% ("GT V") and 23.04% (GT III) (Fig. 2). The latter GT could be separated into two groups based on the tree topology. Division of GT III led to a decrease of intra-genotype heterogeneity: 19.07% (GT IIIa) and 20.11% (GT IIIb). Setting a cut-off at 20.5% nucleotide divergence isolates could still be assigned to genotypes unequivocally (Fig. 3). One isolate from GT IIIb (AF455784 from Kyrgyzstan) showed a particular high divergence (Fig. 1). Animal isolates were distributed in both GTs IIIa and IIIb, GT IV and the "GT V" (Fig. 1).

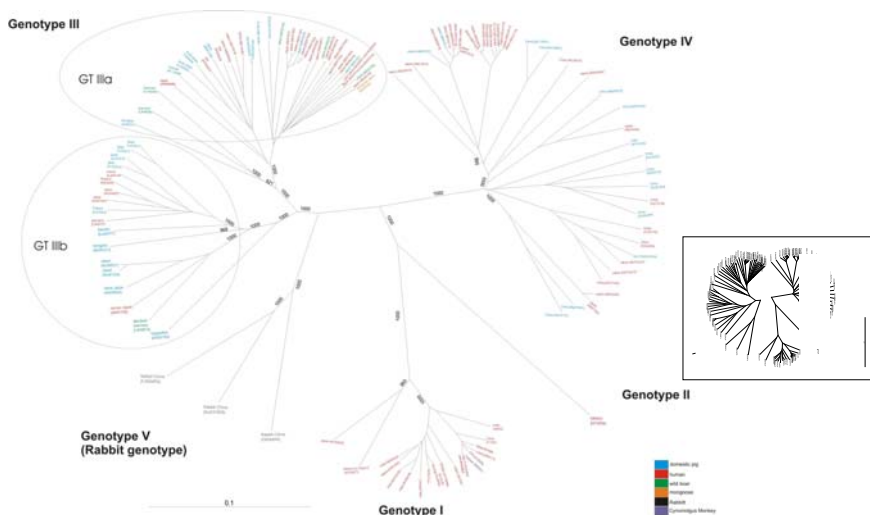


Figure 1: Phylogenetic tree based on 125 complete genomic sequences of HEV isolates. The tree was calculated by the neighbor-joining method. The branch lengths are proportional to the genetic distances. Bootstrap values of 1000 replicates are indicated. Note the color code indicating the species where HEV was isolated. Inset: phylogenetic tree based on maximum-likelihood method showing an almost identical topology.

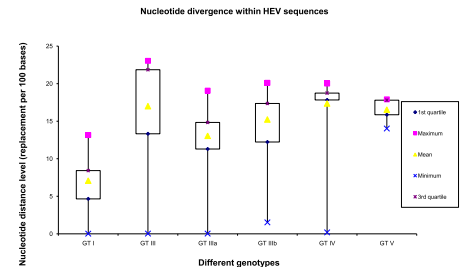


Figure 2: Nucleotide divergence within genotypes I, III and IV. Note the decreasing level of heterogeneity when GT III is split into GT IIIa and GT IIIb.

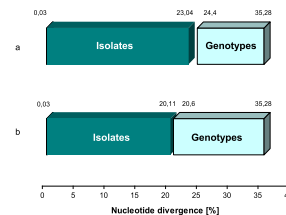


Figure 3: Nucleotide differences between complete HEV sequences at the level of genotypes and isolates. Pairwise comparisons of sequences were performed using Kimura-2 parameter method. a) GTs I, II, III, IV and V b) GTs I, II, IIIa, IIIb, IV and V.

## Discussion and conclusions

Phylogenetic analyses based on complete genomic HEV sequences led to a consistent separation of established genotypes (GTs) and recently discovered isolates from rabbits. High bootstrap values at the lower bifurcations demonstrate the robustness of this phylogeny.

The divergence within GT I is remarkably lower than within genotypes III and IV. So far GT I has only been found in humans. In contrast viruses grouped in GT III and IV have been reported in humans and different animal species. Thus restricted host range may be connected to the lower divergence found within GT I. On the other hand a limited amount of complete genomic sequences were available from GT I and GT II in comparison to GT III and GT IV. This may have biased the analyses.

The separation of GT III into two groups (IIIa and IIIb), is supported by the topology of phylogenetic trees and the calculated genetic distances. The level of heterogeneity within GTs decreased when GT IIIa and IIIb are looked at as separate GTs. It is however still questionable whether these two groups represent separate genotypes.

A separation of GTs into subtypes may be helpful with regard to epidemiological studies. According to our data the phylogenetic grouping at the level of GTs does not coincide with important biological characteristics like virulence or geographical origin. This may in part be due to the lack of information with regard to published sequences. In contrast to GTs IIIa/b and IV the host spectrum of the remaining GTs appears to be restricted to humans (GT I and II) and rabbits (GT "V").

**Conclusion:** The results of our phylogenetic analyses refine the established system of HEV genotypes. Genotype III may be separated into two genetically different groups.

# HYDATIDOSIS IN SUDAN: ARE SHEEP PLAYING A MINOR ROLE IN THE PARASITE CYCLE?

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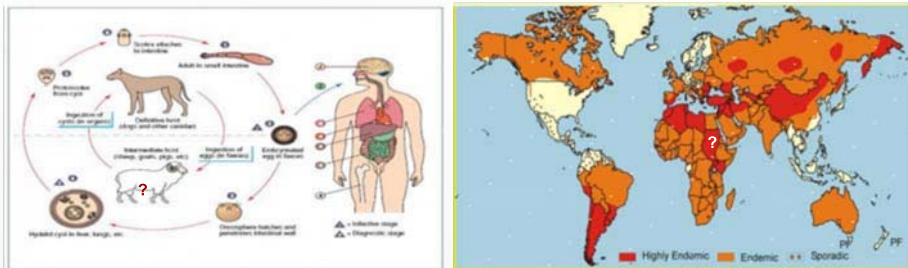


Fig.1 and 2: Life cycle and world-distribution of *Echinococcus granulosus*

## Abstract

This study aimed at the determination of the prevalence, parasitological status and genetic identification of hydatid cysts from sheep in different parts of the Sudan. It was concluded that, sheep play a marginal role in the transmission cycle of the disease in Sudan. This fact is different from data obtained from other regions in Africa as well as parts from southern Sudan, where sheep are heavily involved in the transmission cycle of the disease. Both, the prevalence and fertility rates of the disease in sheep in Western Sudan were higher (11.9% and 19% respectively) comparing to those reported in other investigated areas in Sudan. *E.canadensis* (G6) was identified in all samples and confirmed by mitochondrial gene sequencing of a subset of 15 samples which showed 100% identity with the same strain when compared with data on the GeneBank TM (Accession No. 208063).

## Materials and Methods

### Abattoir Surveys

Multiple visits were made to slaughterhouses in different regions of Sudan (Khartoum, Tamboul and Wad Medani in central Sudan, Nyala in Darfur State (western Sudan), Juba and Malakal in southern Sudan (fig. 3) during the period of May 2001 to July 2003. A total of 10,422 sheep were examined. Lungs, liver, heart, spleen, kidney and peritoneal cavity of slaughtered animals were thoroughly inspected. Prevalence rates, number of cysts /animal, cyst's fertility and predilection sites were determined.

### Genetic characterization

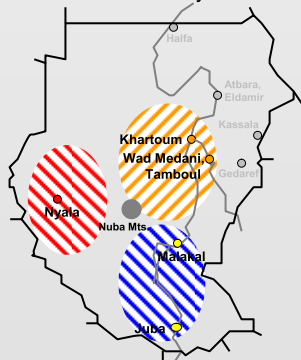
In this survey, we used a previously described PCR system (Dinkel *et al.*, 2004) for species discrimination. As this system does not allow to discriminate between subspecific genotypes (G1/G2/G3, or G6/G7), we also sequenced the partial *cox1* and *nad1* genes of a subset of these samples (Bowles and McManus, 1992).

### Parasitological examination

Cysts were examined macro- and microscopically. Fertility was determined by microscopic detection of protoscolices in aspirated cyst fluid. Sterile or degenerated (calcified or caseated) cysts were classified as infertile. Samples of protoscolices or cyst wall from individual cysts were preserved in 70% ethanol for DNA extraction.

## Results

The results of the study are summarized in table (1) and Figure (4)



Location in Sudan	No	Prevalence [%] (C.I.95%)	Mean No. of cysts / infected animal	Fertility	Predilection site of cysts	No of cysts examined by PCR (fertile-non-fertile)	Species / genotype (n) PCR - Seq.
Central	400	2.5 (1.2-4.4)	1.2	0%	Liver (6/12)	12(0-12)	EC(10) G6(5) NR(2)
Western	9727	11.9 (10.9-12.2)	1.3	19%	Periton cavity (1242/1494)	95 (55-40)	EC (93) G6(5) NR (2)
Southern	295	2.7 (1.2-5.3)	1.0	0%	Liver (8/8)	8 (0-8)	EC (8) G6(5)

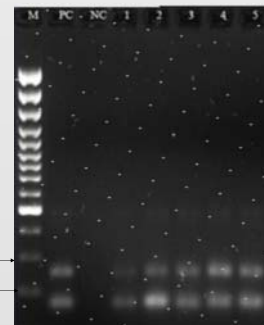


Figure 4: Semi-nested PCR amplification of the 171 bp specific for the camel (G6) strain

Fig. 3: Map of Sudan with locations mentioned in the text. Halifa, Albara and Eldamir are referred to as northern, Khartoum, Wad Medani and Tamboul as central, Kassala and Gedaref as eastern, Nyala as western, Malakal and Juba as southern Sudan.

EC= *E.canadensis* G6/7, NR= No result, PCR: results of species specific PCR system, Suq: mt gene sequencing (*cox1* and *nad1*) for determination of genotype

## Discussion

It is obvious that, sheep play a marginal role in the transmission cycle of the disease in Sudan. This fact is different from data obtained from other regions in Africa as well as parts from southern Sudan, where sheep are heavily involved in the transmission cycle of the disease. This could be attributed to the fact that sheep are usually slaughtered at younger ages before the development of mature cysts. However, in the present study the animals (3-5years) developed calcified or infertile cysts. This may be due to immunoresponce of local Sudanese sheep and goats to hydatidosis infection (Saad and Magzoub, 1989; Omer *et al.*, 2002), or due to the fact that the *E.canadensis* can not express CE in sheep. It may also be concluded that they may have some sort of immunity against *E. Canadensis* as the fertility of the cysts collected from these animals and latterly characterized as *E.canadensis* was very low.

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# An analysis of systemic glucocorticoid use in cats and dogs



**Royal Veterinary College**  
University of London

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## Introduction

Glucocorticoids are among the most widely used (and misused) class of drugs in veterinary medicine (1) yet there is little information on prescribing patterns in general practice. Therapeutic protocols often result from clinical experience, common sense and information from human medicine. However, the adverse metabolic effects are difficult to separate pharmacologically from the therapeutic benefits, making glucocorticoids potent yet potentially dangerous compounds.

The Bateson Independent Inquiry into Dog Breeding (2) recommended that high priority should be given to the creation of a computer-based system for the collection of anonymised diagnoses from veterinary surgeries in order to provide statistically significant prevalence data for each breed. This should build upon the work already started by the Royal Veterinary College.

Description of prescribing practices and analysis of risk factors for treatment with glucocorticoids using computerised clinical records from primary practices could facilitate the benefits while minimising the adverse effects of this important drug category.

## Aims and objectives

1. To describe and compare **prescribing practices** for systemic glucocorticoid pharmacotherapy in **cats and dogs**
2. To evaluate **predictors** for systemic glucocorticoid treatment in primary UK veterinary practice
3. To evaluate **VEctAR Animal Surveillance** (3) as a data collecting system for scientific study analyses

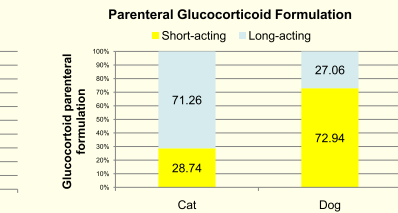
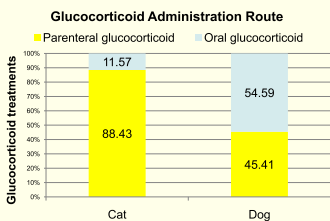
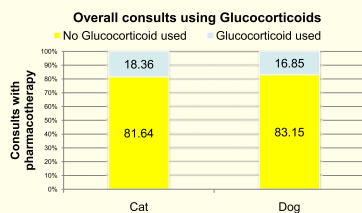
## Materials and Methods

**Data collection:** Three UK small animal practices comprising 7 veterinary clinics were recruited to the **VEctAR Animal Surveillance** pilot project. Practice selection was based on use of a specified computerised Practice Management System (PMS) (RXWorks) as well as a willingness to participate. Vets were asked to assign diagnoses to consults selected from the **VeNom Codes** (4) embedded into their PMS. Data in specified fields on all clinical records from 2007-2009 were captured. The RVC Ethics and Welfare Committee granted ethical approval.

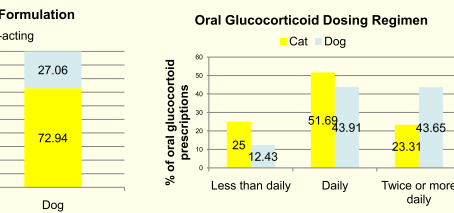
**Analysis:** Extracted data were entered into Microsoft Office Excel 2007 before checking and cleaning. All non-veterinarian and non-cat/non-dog observations were dropped. Summary diagnoses were classified into broad pathophysiological categories as well as on a dermatological basis. Variables assessed included clinic ID, age category, sex, neuter status, purebred status, month, season, dermatological condition, pathophysiological condition, dog-breed size and cat-coat type. Treatment data were searched for systemic glucocorticoid generic and brand names and doses (mg/kg) were calculated using recorded pet weights.

The data were exported to Stata Version 11 for analysis separately by cat and dog based on consultation outcome: whether a systemic glucocorticoid was used or not. Descriptive statistics were generated. Risk factors were analysed using mixed-effects logistic regression. Including 'animal ID' as a random effect took account of clustering of consultations within patients.

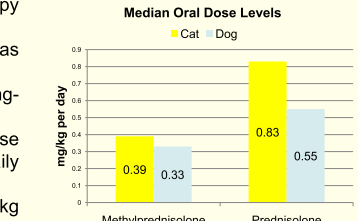
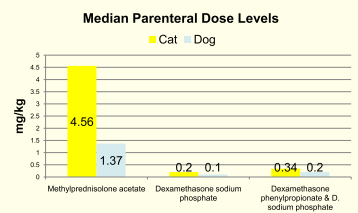
## Descriptive Glucocorticoid Prescribing Practices



**Summary:** Three practices comprising 7 clinics and 24 vets contributed 32,058 consultations overall with pharmacotherapy (cat 11,486, dog 20,572). 56.64% of consults had a summary diagnosis recorded.



- Overall, 17.39% of consultations with pharmacotherapy included systemic glucocorticoids.
- Cats receiving glucocorticoids had 88.43% of treatments as parenteral while dogs had 54.59%.
- Cats received 71.26% of parenteral glucocorticoids as long-acting treatments while dogs had 27.06%.
- For oral glucocorticoids, 51.69% of cats had once daily dose regimens while 43.65% of dogs had twice-or-more daily regimens.
- Median prednisolone daily doses for cats was 0.83mg/kg and for dogs was 0.55mg/kg.



## Risk Factors for Glucocorticoid Therapy

Final multivariable mixed effects regression modelling included clinic ID, pathophysiological indication, skin disease, age category and sex for both cat and dog models. Month of consultation was also included for dogs. Individual clinics varied widely; one clinic had an OR 3.56 (95%CI 2.09-6.09, P<0.0001) for feline consults resulting in glucocorticoid therapy compared with the referent clinic. Neoplastic conditions increased the odds of treatment with glucocorticoids compared with conditions requiring anti-inflammatory (not incl. hypersensitivity conditions) treatment (cat model OR 4.40 95%CI 2.93-6.61, P<0.0001). The odds ratio for glucocorticoid treatment in skin disease cases in dogs was 7.58 (95%CI 6.50-8.84, P<0.0001) compared with non-dermatological cases. Cats aged between 1 and 7 years had 4.13

Variables included	Final Model (P-value)	
	Cat Model	Dog Model
Clinic ID	P<0.0001	P<0.0001
Pathophysiological indication	P<0.0001	P<0.0001
Skin disease	P<0.0001	P<0.0001
Age category	P<0.0001	P<0.0001
Sex	P=0.0027	P=0.0561
Month of consultation	Not Retained	P=0.0622

(95%CI 2.35-7.28, P<0.0001) times the odds of treatment compared with cats under 1 year. Male cats had 0.72 (95%CI 0.56-0.91, P<0.0001) times the odds compared with female cats while male dogs had 1.18 (95%CI 1.00-1.41, P=0.0561) the odds compared with female dogs. The trend was for dog summertime consultations to show a higher odds of glucocorticoid treatment than wintertime.

## VEctAR Animal Surveillance

This study demonstrates that the **VEctAR Animal Surveillance** system can be used to generate meaningful data on a large scale using primary practice caseloads. This data is analysable to answer scientific questions relating to conditions and treatments where primary veterinary practitioners hold essential information. The method of assigning summary diagnoses resulted in a high coding rate and greatly aided analysis and interpretation.

## What next.....

**VEctAR Animal Surveillance** developments since the pilot phase have included:

- ✓Extension of the pilot project to a full national surveillance system
- ✓Recruitment of practice chains, charity, OOH as well as smaller practices
- ✓Continued work with RxWorks and now additional PMS providers
- ✓Ongoing standardisation of veterinary terminology by **The VeNom Coding Group**
- ✓Four PhD studies now incorporating VEctAR Animal Surveillance data

References:  
(1) Glucocorticoids, Mineralocorticoids and Adrenolytic Drugs in Veterinary Pharmacology and Therapeutics. Ferguson, D.C., Dinkola, L., Hoenig, L.Wiley-Blackwell, 2009  
(2) Independent Inquiry into Pedigree Dog Breeding, Bateson, P. Micropress Ltd, Halesworth, Suffolk, 2010  
(3) VEctAR Veterinary Electronic Animal Record Animal Surveillance. [www.rvc.ac.uk/VEctAR](http://www.rvc.ac.uk/VEctAR)  
(4) The Venom Coding Group. [www.venomcoding.org](http://www.venomcoding.org)

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Thank you to RxWorks and selected RxWorks client practices for their collaboration.







# Association of herd BHV-1 seroprevalence with respiratory disease in youngstock of Estonian dairy cattle

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## Introduction

Bovine respiratory disease (BRD) is usually of multifactorial origin, involving infectious, environmental and management-related factors as well as those related to stress and the immunity of the animal. Bovine herpesvirus 1 (BHV-1) is considered to be an important component of the etiological complex causing BRD in cattle.

**Aim** of this study was to investigate the associations of herd bovine herpesvirus 1 (BHV-1) status and respiratory disease (BRD) occurrence in dairy calves and pre-breeding heifers. In addition, associations between farm management related factors and the occurrence of BRD in that age group was evaluated.

## Methods

Serum samples were collected from cows and youngstock from 103 dairy cattle herds and analyzed for antibodies against BHV-1, bovine respiratory syncytial virus (BRSV), bovine virus diarrhea virus (BVDV), and *Mycoplasma bovis* (*M. bovis*). Questionnaire was used to register herd management practices and disease occurrence.

## Results

Table 1. Results of logistic regression analysis for risk factors for high occurrence of respiratory disease in calves until three months old (99 herds)

Risk factor	Herds (n)	OR	p	95% CI
<b>BHV-1 prevalence in cows<sup>a</sup></b>				
0	38	1	-	-
1-49%	26	14.8	0.005	2.3; 95.5
>50%	35	19.2	0.002	3.0; 121.8
<b>BVDV in heifers</b>				
0	75	1	-	-
1	24	5.1	0.020	1.3; 20.1
<b>Herd size<sup>b</sup></b>				
20-99 cows	40	1	-	-
100-199 cows	18	1.7	0.557	0.3; 9.3
200-399 cows	23	8.0	0.008	1.7; 37.3
>400 cows	18	6.4	0.029	1.2; 33.8

Table 2. Results of logistic regression analysis for risk factors for high occurrence of respiratory disease in heifers three to sixteen months old (99 herds)

Risk factor	Herds (n)	OR	p	95% CI
<b>BVDV in heifers</b>				
0	76	1	-	-
1	23	4.3	0.027	1.2; 15.8
<b>Herd size</b>				
20-99	40	1	-	-
100-199	19	4.9	0.113	0.7; 34.4
200-399	23	5.3	0.065	0.9; 31.6
>400	17	8.1	0.022	1.4; 49.1

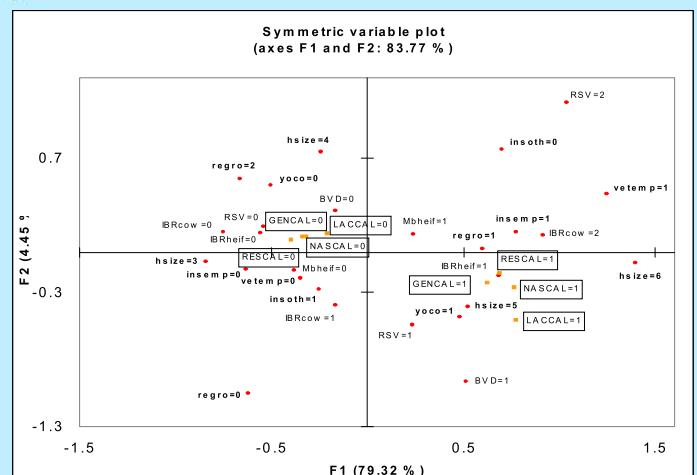


Figure 1. Graphical display of Multiple Correspondence Analysis, with respect to axis 1 and 2 for high occurrence of respiratory disease symptoms in calves until three months old (99 herds)

## Conclusions

- BHV-1 and BVDV were related to BRD in unweaned calves
- BVDV is associated with BRD in older youngstock
- Higher occurrence of respiratory disease is generally more likely in larger herds
- On-farm employees can participate in spreading of the disease
- Holding youngstock separately from cows until pregnancy and purchasing animals were factors related to a higher risk for BRD

## ACKNOWLEDGEMENTS

This research was supported financially by the Estonian Ministry of Agriculture (Research contract 34-23 2006-2008) and Estonian Ministry of Education (Research project 8-2/T9001).

# Animal Production Systems in the Kaski district of Nepal

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## Introduction

Agriculture of developing countries is predominately focused on small farms, which hold great potential for increasing prosperity of communities. Such farms, however, are usually resource-limited and farming and livestock husbandry occur in the context of a hand-to-mouth subsistence living. Implementation of solutions to improve productivity need to take into account local situations and available resources, while not imposing prohibitive costs on farmers.

Nepal is among the poorest and least developed countries in the world, with almost ¼ of its population living below the poverty line. Agriculture is the mainstay of the economy, with the majority of producers being from small farms with low-producing animals. We surveyed 85 farms in the rural Kaski district with the goal of describing animal production systems and determining areas where effective interventions can be made to increase animal health and productivity.

## Methods

85 randomly selected farms from the Kaski district were visited by the research team consisting of a Nepali veterinarian and 3 students from the University of Pennsylvania, School of Veterinary Medicine. Farmers were interviewed with a questionnaire that was developed by the research team and pre-tested. Animals were evaluated for body condition and their weights determined with weight-tapes. Samples of forage and concentrates were taken from a sub-sample of farms and analyzed for energy, protein, soluble fibers and minerals contents. CPM Dairy was used to determine production levels obtainable from rations fed to animals with local feeds.

## Results

### Demographics:

Animals included buffaloes, goats, and cows. 13% of farms had all three species, while 50% had goats and buffaloes.

	Total number/farm	# pre-pubertal	#pregnant	#producing	Breed
Buffalo	2.83	1.05	1.19	0.70	84% local/parkote 16% Murrah cross
Cattle	2.76	0.63	0.47	0.74	95% local breed 5% Jersey cross
Goats	5.17	2.1	1.2	N/A	10% Jamanapari x 88% Local (khar/sinhal) 1% Sanen x Austr. Boer

### Reproduction

	Age of 1 <sup>st</sup> breeding, mean(SD)	Average # of breeding attempts, mean(SD)	Dry period (months), mean(SD)	Calving/Kidding interval (months), mean(SD)	parturition number, mean(SD)
Buffalo	3.2 years (0.41)	1.6 (0.53)	3.4 (1.8)	12.9 (2.1)	12.1 (2.5)
Cows	3.1 years (-.54)	1.6 (0.55)	4.4 (1.5)	13.4 (4.0)	11.0 (2.3)
Goats	5.5 months (1.6)	1.6 (0.54)	1.6 (0.69)	6.4 (0.69)	10.9 (1.7)

### Housing and husbandry

- Housing generally consisted of open stalls (Fig 1).
- 44% of water buffalo, 92% of cattle and 50% of goats had access to pastures, usually for a minimum of 8 hours a day.
- Anthelmintics were administered regularly, while vaccination was mostly done on an as-needed basis
- Weaning and artificial insemination were not performed.
- 42% of water buffalo farmers sold milk, 8.3% of cattle farmers sold milk and 100% of goat farmers sold goats for meat.

### Nutrition

- 78% of buffalo, 67% of cattle and 37% of goats were fed concentrates. (wheat bran, rice bran, corn flour, millet flour) (Fig 2c).
- Vitamin and mineral supplementation was infrequent
- Most farms made straw (Fig 2b)
- Feed costs averaged \$45/buffalo/year, \$15/cow/yr, \$8/goat/year.
- Forage and concentrate nutritional content was insufficient to support increased production.

### Conclusions

- Farms are small and often self-sufficient producers.
- Increased income could be generated from increasing and selling more animal products.
- Nutrition is likely the limiting factor for increasing production.
- Community Animal Health Workers have a significant role to play in improving animal health in rural areas underserved by veterinarians.



Fig 1a: Open goat stalls



Fig 1b: Water buffalo stalls



Fig 1c: Raised goat housing



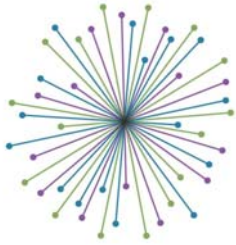
Fig 2a: Fresh greens



Fig 2b: Straw-making



Fig 2c: Corn grinding



CENTRE FOR EVIDENCE-BASED  
VETERINARY MEDICINE  
Putting research into practice

# Consultations in small animal practice: Does reaching a diagnosis affect outcome?

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**What is a diagnosis?:** "A diagnosis is the label given to a disease with certain clinical or pathologic characteristics applicable to a particular case" (Radostits et al, 2000). This is not always easy to define, for example:

- **Congestive Heart Failure:** Is this a diagnosis? Or can we only reach a diagnosis if we know the inciting cause e.g. Dilated Cardiomyopathy?
- **Otitis externa:** Again, is this a diagnosis in its own right? Or must we know if it is secondary to atopy, ear mites, ear conformation etc?
- **Atopy:** When does it become appropriate to treat for atopy in a pruritic dog? Based on consistent history, clinical signs, signalment and skin lesions? After exclusion of all other causes? Only after specific allergen testing? Or variable depending on the case?

**Aims:** The aim of this study was to conduct practice-based research in order to collect data on commonly encountered conditions and presentations in small animal practice. In particular, the aim was to look at the type of diagnosis made and whether this impacted upon the outcome of the consultation

## Methods:

- Development of diagnosis definitions, a data collection form and a network of 8 sentinel practices
- Collection of data by direct observation of consultations
- Data recorded for all presenting complaints as well as additional problems raised by the owner or vet during the consultation
- Each problem discussed was assigned only one diagnosis type but could be assigned multiple outcome types if necessary e.g. Treat and Manage
- Pilot study complete: single day of data collection at each sentinel (n=8)
- Future plan: collection of data for three separate one week periods at each practice over a 1-2 year period

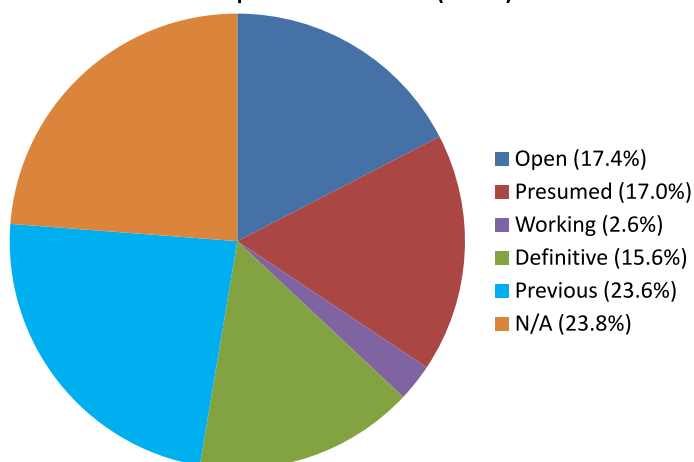
## Definitions of Diagnosis Type:

- **Open Diagnosis:** No single diagnosis is significantly more likely, multiple differentials are being considered at the conclusion of the consult
- **Presumed Diagnosis:** A diagnosis based on minimal evidence/clinical suspicion, upon which therapy or other non-diagnostic interventions may be based
- **Working Diagnosis:** A diagnosis based on early confirmatory evidence, allowing early management of the disease while awaiting confirmation from the results of further diagnostic tests
- **Definitive Diagnosis:** A diagnosis with a high level of confidence, where any necessary diagnostic s including 'gold standard' tests have been completed
- **Previous Diagnosis:** A diagnosis made during a previous consultation or diagnostic work-up
- **N/A:** A diagnosis is not applicable as the problem or 'complaint' being discussed relates to preventive medicine

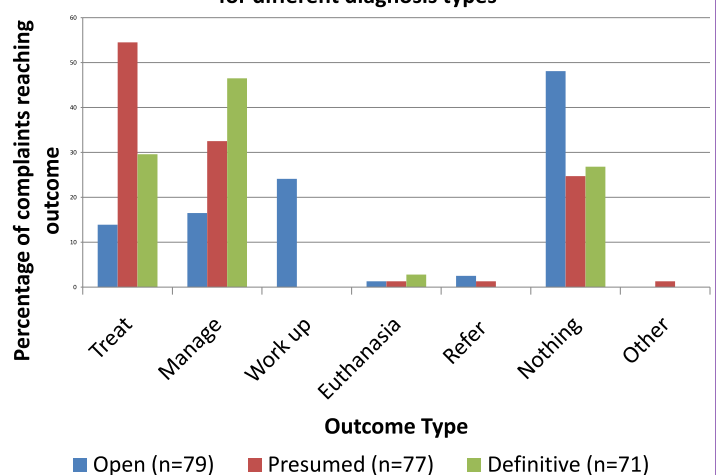
## Results:

Data collected from 181 consultations involving 199 animals with 454 separate problems, during a single day at each of the 8 sentinel practices

Pie chart showing frequency of diagnosis types reached for all problems discussed (n=454)



Bar chart showing frequency of consultation outcomes for different diagnosis types



**Conclusions:** The initial results from this pilot study suggest that definitive diagnoses are made relatively infrequently in the consult room, while vets spend almost a quarter of their time dealing with previously diagnosed cases. Comparison of diagnosis and outcome types suggests that the consultation outcome seems to vary considerably depending upon the diagnosis made. Perhaps unsurprisingly an open diagnosis is more likely to result in a diagnostic work up, however it is also more likely to result in the decision to do nothing, and less likely to result in therapeutic treatment or management compared with presumed or definitive diagnoses. However, further investigation is needed to make more meaningful conclusions from this and to determine what other factors influence the likelihood of a particular diagnosis or outcome type. Diagnosis is a difficult term to define and the consideration of different types or levels of diagnosis may be useful when interpreting data from first opinion practice.

**Acknowledgements:** Thanks very much to the sentinel practices and their clients for their involvement in the study, and to Novartis Animal Health and University of Nottingham for sponsoring this research

## References:

Radostits, O.M. Et al (2000). "Making a diagnosis" In: Veterinary Clinical Examination and Diagnosis, W.B. Saunders, London, p11-52

# Simulation of the seasonal cycles of bird, equine and human West Nile virus cases

Franz Rubel and Katharina Brugger

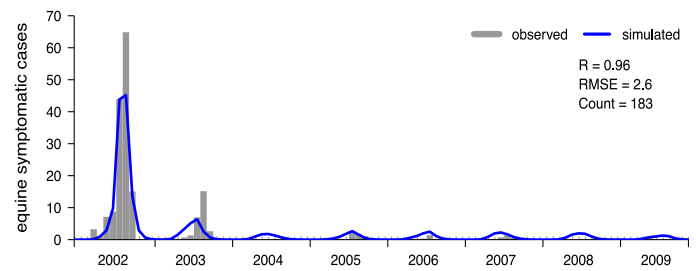
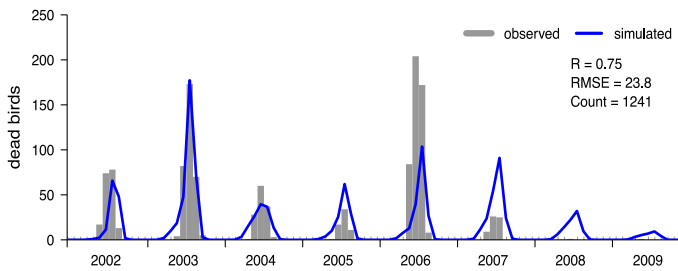
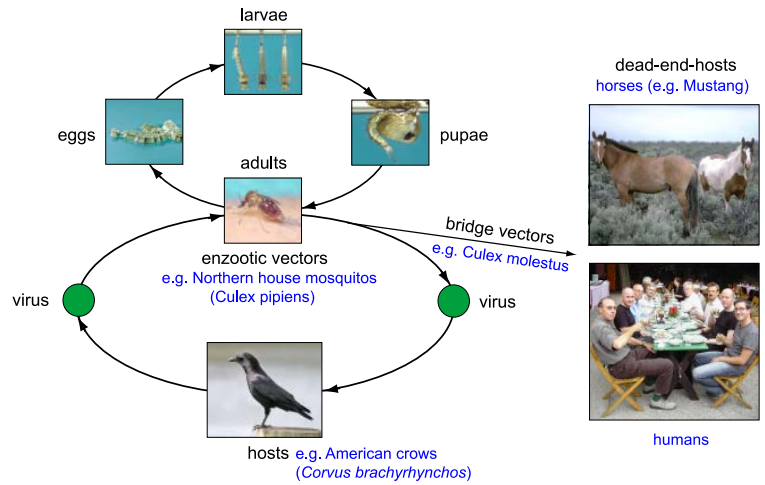
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## Motivation

The West Nile virus (WNV) is an arbovirus circulating in a natural transmission cycle between mosquitoes (enzootic vectors) and birds (amplifying hosts). Additionally, mainly horses and humans (dead-end hosts) may be infected by blood-feeding mosquitoes (bridge vectors). We developed an epidemic model for the simulation of the WNV dynamics of birds, horses and humans in the U.S., which we apply to the Minneapolis metropolitan area (Minnesota).

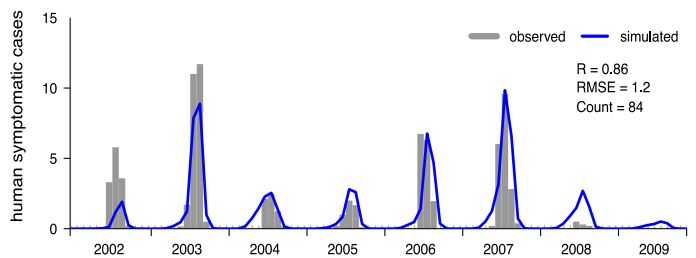
## Method

Our SEIR-type model comprises a total of 19 compartments, that are 4 compartments for mosquitoes and 5 compartments or health states for each of the 3 host species. It is the first WNV model that simulates the seasonal cycle by explicitly considering the environmental temperature (see the theoretical background at the bottom of the poster where temperature dependent parameters are marked in red). Generally, all parameters were estimated from literature or census. Special features of the epidemic model are: (1) density dependent population dynamics of wild birds and mosquito larvae, (2) temperature dependent mosquito parameters, including biting rate, hibernation and extrinsic incubation period, (4) population dynamics of horses and humans following national census, and (5) frequency dependent virus transmission (Laperriere *et al.*, 2011).



## Results

We adjusted our WNV model to fit monthly totals of reported bird, equine and human cases. From this process we estimated that the proportion of actually WNV-induced dead birds reported by the Centers for Disease Control and Prevention is about 0.8%, whereas 7.3% of equine and 10.7% of human cases were reported. This is consistent with referenced expert opinions whereby about 10% of equine and human cases are symptomatic (the other 90% of asymptomatic cases are usually not reported). Despite the restricted completeness of surveillance data, all major peaks in the observed time series were caught by the simulations. Correlations between observed and simulated time series were  $R = 0.75$  for dead birds,  $R = 0.96$  for symptomatic equine cases and  $R = 0.86$  for human neuroinvasive cases. Our WNV model may also be applied to other arbovirus epidemics. For example, Rubel *et al.* (2008) and Rubel and Brugger (2009) applied it to explain the Usutu virus epidemics in Vienna, Austria.

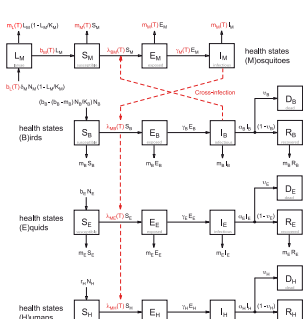


## References

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- Brugger, K., and F. Rubel, 2009: Simulation of climate-change scenarios to explain Usutu-virus dynamics in Austria. *Prev. Vet. Med.*, **88**, 24-31.
- Rubel, F., *et al.*, 2008: Explaining Usutu virus dynamics in Austria: Model development and calibration, *Prev. Vet. Med.*, **85**, 166-186.

### Theoretical background

#### Epidemic Model



#### Equations

$$\frac{dL_{M1}}{dt} = (b_L(T) \delta_{L_{M1}} N_{L_{M1}} - m_L(T) L_{M1}) \left(1 - \frac{L_{M1}}{K_{M1}}\right) - b_{M1}(T) L_{M1}$$

$$\frac{dL_{M2}}{dt} = -\lambda_{L_{M2}}(T) S_{M1} + b_{M1}(T) L_{M1} - m_{L_{M2}}(T) L_{M2}$$

$$\frac{dL_{M3}}{dt} = \lambda_{L_{M3}}(T) S_{M1} - \gamma_{L_{M3}}(T) L_{M3} - m_{L_{M3}}(T) L_{M3}$$

$$\frac{dL_{M4}}{dt} = \gamma_{L_{M4}}(T) L_{M3} - m_{L_{M4}}(T) L_{M4}$$

$$\frac{dS_{H1}}{dt} = (b_H - (b_H - m_H) \frac{N_{H1}}{K_{H1}}) N_{H1} - \lambda_{M4}(T) S_{H1} - m_{H1} S_{H1}$$

$$\frac{dE_{H1}}{dt} = \lambda_{M4}(T) S_{H1} - \gamma_{E_{H1}} E_{H1} - m_{H1} E_{H1}$$

$$\frac{dI_{H1}}{dt} = \gamma_{E_{H1}} E_{H1} - \alpha_{H1} I_{H1} - m_{H1} I_{H1}$$

$$\frac{dR_{H1}}{dt} = (1 - \alpha_{H1}) \alpha_{H1} I_{H1} - m_{H1} R_{H1}$$

$$\frac{dD_{H1}}{dt} = \alpha_{H1} \alpha_{H1} I_{H1}$$

$$\frac{dS_E}{dt} = b_E N_E - \lambda_{M4}(T) S_E - m_E S_E$$

$$\frac{dE_E}{dt} = \lambda_{M4}(T) S_E - \gamma_{E_E} E_E - m_E E_E$$

$$\frac{dI_E}{dt} = \gamma_{E_E} E_E - \alpha_{E1} I_E - m_E I_E$$

$$\frac{dR_E}{dt} = (1 - \alpha_{E1}) \alpha_{E1} I_E - m_E R_E$$

$$\frac{dD_E}{dt} = \alpha_{E1} \alpha_{E1} I_E$$

$$\frac{dS_H}{dt} = r_H N_H - \lambda_{M4}(T) S_H$$

$$\frac{dE_H}{dt} = \lambda_{M4}(T) S_H - \gamma_{H} E_H$$

$$\frac{dI_H}{dt} = \gamma_{H} E_H - \alpha_{H1} I_H$$

$$\frac{dR_H}{dt} = (1 - \alpha_{H1}) \alpha_{H1} I_H$$

$$\frac{dD_H}{dt} = \alpha_{H1} \alpha_{H1} I_H$$

#### Parameters

Param.	Value	Interpretation	Param.	Value	Interpretation
$b_L$	$f(T)$	Birth rate, larvae	$m_E$	0.00034	Mortality rate, equids
$m_L$	$f(T)$	Mortality rate, larvae	$\alpha_{E1}$	0.4	Fraction birds dying due to infection
$b_M$	$f(T)$	Birth rate, mosquitoes	$\gamma_{H}$	0.25	Rate with $1/\gamma_H$
$m_M$	$f(T)$	Mortality rate, mosquitoes	$\alpha_{H1}$	0.004	Fraction humans dying due to infection
$p_{M1}$	1.0	Transmission probability by infectious mosquitoes	$\phi_B$	0.03	Mosquito-to-bird ratio
$\gamma_{M1}$	$f(T)$	Rate with $1/\gamma_{M1}$	$b_H$	0.000555	Birth rate, humans
$\delta_{M1}$	$f(T)$	Rate with $1/\delta_{M1}$	$m_H$	0.00034	Mortality rate, humans
$\lambda_{M4}$	$f(T)$	Fraction mosquitoes non-hibernating	$\alpha_{H1}$	0.5	Removal rate, humans
$b_E$	0.00016	Birth rate, equids	$\phi_B$	0.03	Mosquito-to-bird ratio
$m_E$	0.00011	Mortality rate, equids	$\phi_B$	0.03	Mosquito-to-bird ratio
$\alpha_{E1}$	0.2	Removal rate, equids	$\phi_B$	0.03	Mosquito-to-bird ratio
$\gamma_E$	0.05	Rate with $1/\gamma_E$	$\phi_B$	0.03	Mosquito-to-bird ratio
$\mu_E$	0.04	Fraction equids dying due to infection	$\phi_B$	0.03	Mosquito-to-bird ratio
$\phi_E$	300	Mosquito-to-equid ratio	$\phi_B$	0.03	Mosquito-to-bird ratio

$$\text{Forces of Infection } \lambda_{M4}(T) = \delta_{M4}(T) \frac{I_B}{K_B} + \lambda_{M4}(T) \frac{I_H}{K_H} \phi_B \phi_B \dots$$

# Modelling Footrot: A Work in Progress

by Vinca Russell<sup>1,2</sup>, Laura Green<sup>1</sup>, Stephen Bishop<sup>2</sup> and Graham Medley<sup>1</sup>

<sup>1</sup>University of Warwick and <sup>2</sup>The Roslin Institute (University of Edinburgh)

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## Introduction

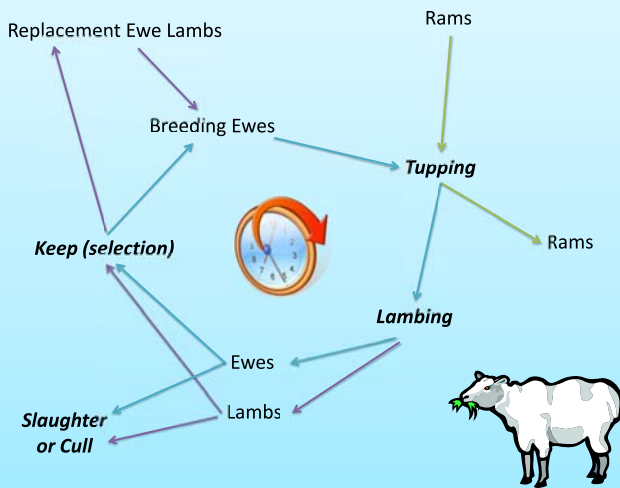
**Footrot** is an infectious bacterial disease of sheep transmitted between animals via contaminated pasture. The primary aetiological agent of footrot is *Dichelobacter nodosus* and its clinical signs are foot lesions and lameness. Footrot also results in a decrease in production traits such as live-weight of lambs and is a serious welfare concern. Although it can be treated with antibiotics, these treatments are time-consuming, costly and provide only temporary relief. There is no long term immunity to footrot and it is common for sheep to become infected multiple times.

**Simulation models** allow us to explore long term consequences of interventions and breeding programmes on disease patterns. We have constructed a stochastic simulation model that mimics as closely as possible the behaviours seen in real sheep flocks, including life cycles, disease patterns, bacteria in the environment and heritable genetic characteristics (e.g. susceptibility and recoverability). The model will be validated using a large longitudinal dataset containing a series of measurements of lameness and lesions in a sheep flock including both lambs and ewes.

The **model population** is made up of 200 ewes aged 0-5 years. Each year ewes have 0-3 lambs that inherit maternal values for susceptibility. All ewes aged 5 are culled, along with 5% of other adult ewes, all male lambs and any female lambs that are not required for restocking. Lambs are born in March and culled at the end of August. Rams will be introduced for tupping only but at present are not included. The model runs for a simulated time period of 20 years, incorporating 20 generations of sheep.

## Life Cycle

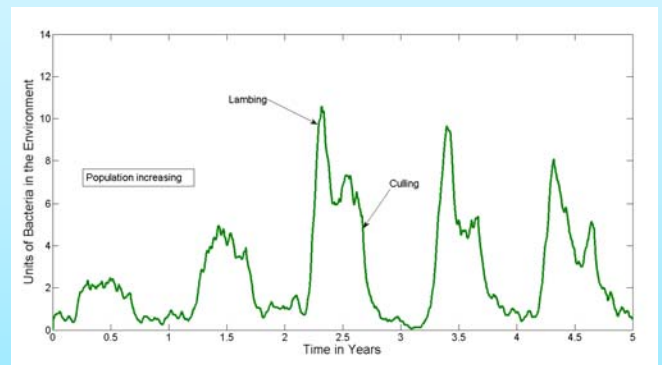
• Model includes the sheep life cycle as indicated in the diagram below. Key events are in bold.



## Bacteria

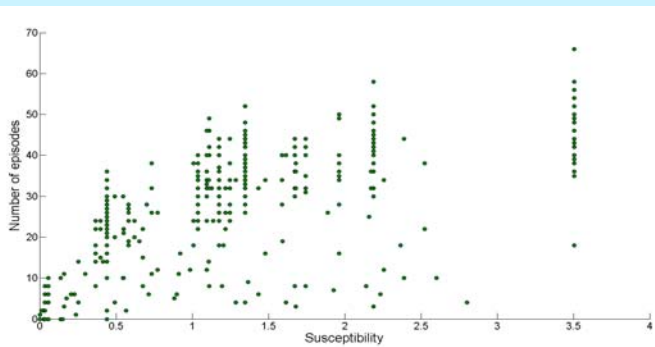
- Bacteria shed from sheep onto pasture.
- Bacteria transmitted from pasture to sheep.
- Bacteria die in the environment after time (average 7 day survival).

The graph below shows levels of bacteria in the environment over time from the model. Major peaks and troughs coincide with lambing and culling events.



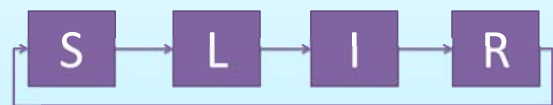
## Host Genetics

- Different sheep can have different levels of resistance to disease.
- Resistance may be measured in different ways:
  - Susceptibility to initial infection.
  - Recoverability – the time taken to recover from disease.
  - Revertability – the time taken to lose immunity and revert to a susceptible state.
- The graph below shows the number of disease episodes for five-year old sheep with different susceptibilities.

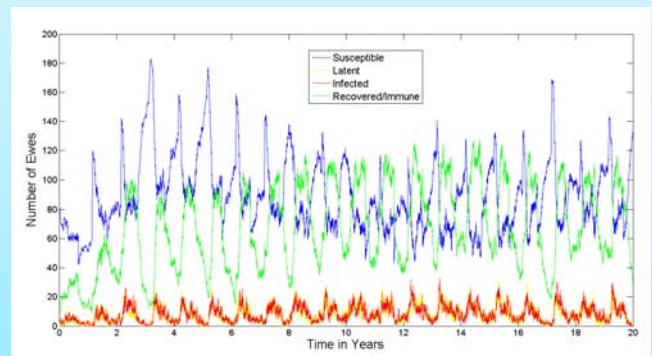


## Disease

- Modelled using an S-L-I-R-S (susceptible, latent, infectious, recovered) model:



- Disease patterns from the model are shown in the graph below.



## What's next?

- Interrogation of the model to examine the interactions between genetics and epidemiology.
- Inclusion of interventions and selective breeding to see what effects these have on disease patterns.

## Cattle

# Quantification of transmission ( $R_0$ ) of bluetongue virus serotype-8 within dairy herds based on serological field data

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## Aim of the study

The goal of the study was to determine the basic reproduction rate ( $R_0$ ) of BTv-8 between Dutch dairy cattle in the 2007 BTv-8 epidemic.

## Material & Methods

### Data

In 2007, a sentinel network was initiated to monitor the circulation of BTv-8 in the Netherlands. In May, 275 dairy herds were selected across the Netherlands and seronegative cows were tested each month for antibodies in their milk. The percentage of seroconversions per herd per month was derived (Figure 1) the  $R_0$  of the infection could be determined.

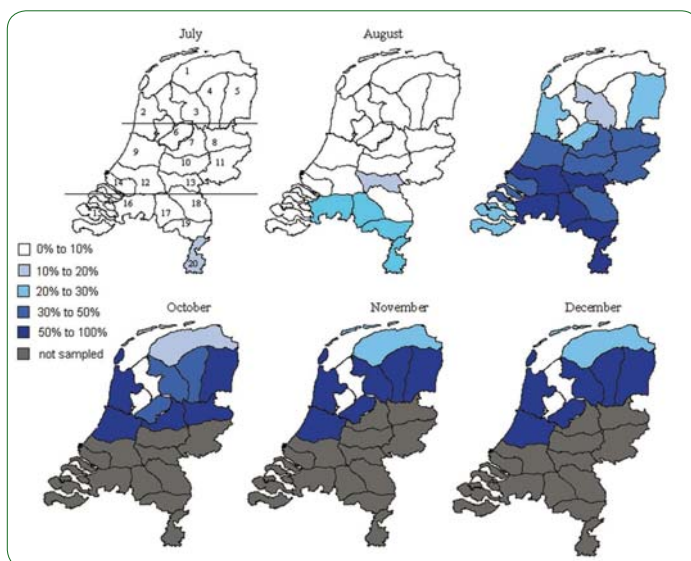


Figure 1. Average within-herd BTv-8 seroprevalence in the Netherlands. Horizontal lines in the first map represent the region North, Central and South.

### Model

The cattle population per time  $t$  was divided into three subclasses of cows that were susceptible, infectious or recovered. The proportion recovered cows was derived from the field data as the proportion of seropositive cows for each dairy herd per measurement at time  $t$ . These seropositive cows became seropositive after an infectious period which was caused by being bitten by an BTv-8 infectious vector (Figure 2).

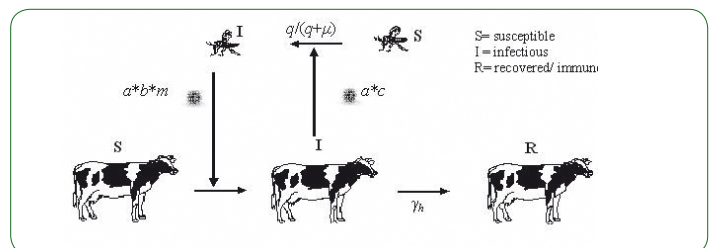


Figure 2. Description of the transmission route of BTv-8.

- $a$  biting rate per day
- $b$  effectiveness transmission from vector to host
- $c$  effectiveness transmission from host to vector
- $m$  number of *Culicoides* per day
- $Y_h$  recovery rate per day
- $(q/(q+\mu))$  P to survive the incubation period  $q$  is P to leave the incubation state and  $\mu$  is the mortality rate

### Results

The mean between-cattle  $R_0$  in the Netherlands in 2007 was 3.8 (95% CI: 3.5-4.1). Overall median  $R_0$  values were highest between August and October (3.5). However, per region (south, central or north) the month in which the  $R_0$  value was highest differed (Figure 3).

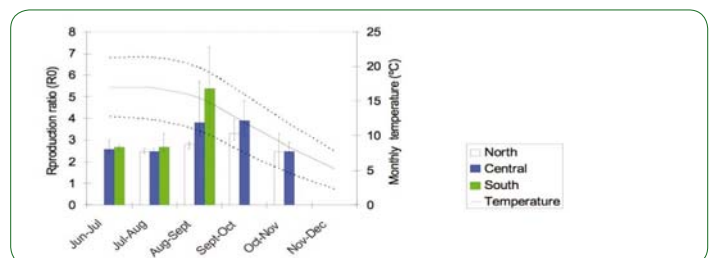


Figure 3. Median and inter quartile range of  $R_0$  per month from June until December 2007 for region north, central and south within Dutch dairy herds.

### Conclusion

The transmission of BTv-8 is highly dependent on outside temperature. Nevertheless, during the 2007 BTv-8 epidemic in the Netherlands, one BTv-8 infectious cow could infect on average 3.8 other cattle by bites of *Culicoides*. This  $R_0$  seemed to represent the within-herd spread of BTv-8 in the field well and these transmission rates could apply to other countries in which BTv-8 emerges, given a similar climate, grazing patterns and barn types as in the Netherlands.

### Acknowledgements

This study was financially supported by the product board for Livestock, Meat and Eggs (PVE) and the ministry of Agriculture, Nature and Food Quality (LNV).

# Epidemiological Investigations on Bovine Neonatal Pancytopenia in Young Calves

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## Introduction

Since 2006 an increased incidence of haemorrhagic diathesis in young calves has been observed in Germany and other European countries (Belgium, France, Hungary, Italy, Ireland, the Netherlands, Spain, UK), while it was never observed in Austria, Switzerland, or Denmark. The exact cause of the disease is still unknown. Some calves are noticed by their spontaneous bleedings of the skin or blood in the faeces, while others are treated due to neonatal disorders or disturbance of the general condition. Despite blood transfusions, most affected calves die or have to be euthanised.

## Material and Methods

The Clinic for Ruminants has been involved in the research on BNP since 2007, and many cases have been presented to the clinic. From other animals blood samples were sent by the local veterinarians to the clinic if they had the suspicion of BNP. Post-mortem reports of some of these animals were also sent to the clinic.

Information on the animal and the farm of origin was asked via telephone and recorded on a questionnaire. Analysis of blood values is undertaken.

The diagnosis Bovine Neonatal Pancytopenia was based on following criteria:

- Multiple haemorrhages (blood in the faeces, skin bleedings, petechiae) (fig. 1 and fig.2)
- Age up to 4 weeks
- Thrombocytopenia ( $< 200\ 000/\mu$ ) and leucocytopenia ( $< 4000/\mu$ l)
- No indications of septicaemia
- Bone marrow depletion (Panmyelophthisis)



Figure 1: Skin bleeding of a BNP calf



Figure 2: Petechiae in the mucosa

In a further study, a one-page questionnaire was mailed to all large animal veterinary practices in Bavaria (about 1000) in July, 2009, and July, 2010. Veterinarians were asked to state if they had observed the diseases in their practice and if so, at what time and in how many farms. The addresses of the veterinarians were provided by the Bavarian veterinary chamber. The questionnaire was also published in the German Veterinary Journal ('Deutsches Tierärzteblatt').

## Discussion and Conclusion

Although the animals were presented on average at the age of 14 days, it has to be assumed that the disease has been present for some time prior to that already.

The number of affected calves per farm is most likely severely underreported, as not all cases are identified or confirmed, as there are most likely also subclinical cases, and due to the bad prognosis for affected calves new cases might not be reported again.

The link with a particular BVDV vaccine is getting stronger, although the exact mechanism is still unknown. However, this vaccine has been withdrawn from the markets in Europe in June, 2010.

## Results

Up to the beginning of March 2011 in total 459 confirmed cases of BNP of 284 different farms were reported to the clinic of ruminants.

The gender of 308 calves was known, whereby 46% were male and 54% female.

Affected calves were between 2 and 28 days old, on average 14 days (fig. 3).

Several breeds were affected; most cases were of German Simmental (72%) followed by Holstein Friesians with 15%. However, Red Holsteins, Brown Swiss and cross breeds were also affected.

There is great variation in the number of calves being affected between farms – mostly only individual cases are reported, but in some farms up to 20% of their calves are affected. Also, numerous cows are reported, that have had affected calves for several consecutive years (up to four affected calves).

The date when the BNP cases were reported is shown in figure 4.

There is increasing evidence that the vaccination against BVDV is linked to the disease – the dams of 440 of 459 affected calves were vaccinated with one particular BVDV-vaccine.

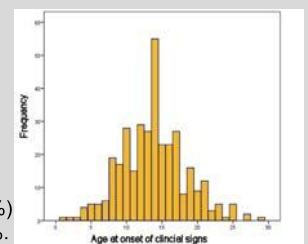


Figure 3: Age of affected calves

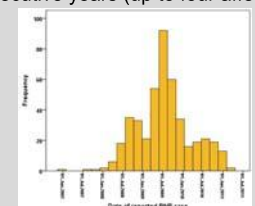


Figure 4: Date of BNP cases

In the questionnaire survey of large animal practices, in total 419 veterinary practices (that were servicing cattle farms) responded to the questionnaire in 2009, while it were only 199 in 2010. In 2009 87 (20.8%) veterinarians stated, that they have observed the disease on the farms of their clients.

Of 349 veterinary practices, the zip code was stated, of which 305 were in Bavaria. The regional distribution of these practices is shown in figure 5.

No regional clustering could be detected.

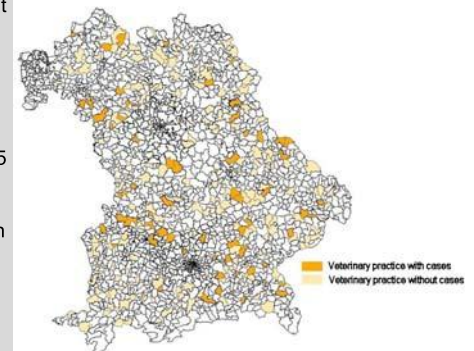


Figure 5: Geographical distribution of practices with and without BNP cases

## Acknowledgements

Thanks to all veterinarians and farm managers for their support.



# A quantitative assessment of the risk of pig-human transmission of pandemic (H1N1) 2009 swine influenza from exposure on pig holdings in Great Britain



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- (3) Combating Swine Influenza Initiative (COSI) consortium, c/o VLA Weybridge



**Introduction:** H1N1 is a subtype of the influenza A virus that has a wide host range and is commonly found in pigs. The virus mutated into the novel pandemic strain pH1N1 which contains swine, avian and human genetic elements. The virus first emerged in Mexico in March 2009 and began to cause illness in the United Kingdom about a month later. While the majority of human pH1N1 infections are due to human-human transmission, a number of studies have indicated that some people who work in the pig industry have been exposed to virus from infected pigs. We present a quantitative risk assessment (QRA) to investigate the risk posed to humans from exposure to pigs infected with pH1N1 on farms in Great Britain (GB).

## Method:

- The QRA is based on a quantitative, stochastic, Monte-Carlo simulation model for transmission of virus from pigs to humans within a farm, with inputs from a between-herd network model, based on observed pig movement records. The overall model framework is shown in Figure 1 and is divided into three components comprising release, exposure and consequence assessments.
- The release assessment uses results from a between-herd network model to estimate the number of pig herds infected with pH1N1 in GB over one year and the length of time that they will remain infected. An important factor in the release is the initial "seeder" farm (i.e. the first farm that was infected), specifically whether it is a smallholding or a commercial (professional) producer. Each iteration of the model predicts the consequence of a single release of infection in to the pig population and subsequent between-herd transmission; it does not consider multiple "seeds" nor the likelihood that any such "seed" will occur.
- The exposure assessment estimates the number of people who work on or visit an infected pig farm (categorised by whether they are staff or visitors) and whether they will be exposed to pH1N1. We estimate exposure via direct and indirect contact with pigs. Exposure via direct contact is assumed to occur only when people are inside the pig buildings; otherwise exposure is via indirect contact.
- The consequence assessment then estimates whether the exposed individuals will become infected, based on an estimate of the attack rate, i.e. the proportion of individuals exposed to pH1N1 (2009) that will become infected, split up by staff and visitor types.
- While the model framework is based on a number of assumptions that affect confidence in the accuracy of point estimates, it's strength is in analysing the relative differences in risk of infection due to factors such as farm type, where the infection was seeded and intervention measures. The model is designed to be able to incorporate new evidence and can highlight areas where future research would be beneficial.

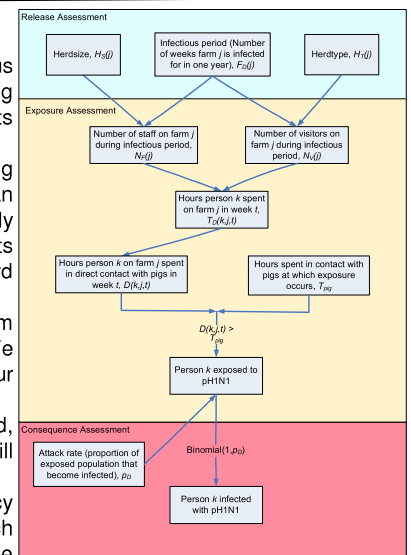


Figure 1: Risk assessment framework

## Results:

- Figure 2 shows the relative effect of farm type and staff/visitor type, over all iterations (each iteration represents a possible realisation of the infection that could happen over one year), on the number of people infected on a farm with infected pigs. We can see that staff from large commercial holdings are by far the most likely to be infected.
- Further analysis estimated that about 67% of small holdings with infected pigs had no infected staff over the course of a year, whereas this figure was only about 40% for large commercial farms. There was a similar pattern for the visitors, suggesting that infection in pigs in larger holdings is more likely to result in pig-human infection.
- Figure 3 shows relative effect of the initial seed farm on the annual number of human infections. There is a clear positive correlation between the size of the initial farm and the risk of human infections.
- Figure 4 shows the effect of the scenario analysis. We vary input parameters by increasing and decreasing them by 10%, 50% and 90% and measure the percentage change in the number of human infections of pH1N1, compared to the baseline model. The results suggest that an intervention that reduces the length of time infection is present on the farm, or the attack rate, could lead to a significant reduction in human infections. The scenario where individuals with 2 hours direct contact with pigs became exposed ( $T_{pig}=2$ ) produced a disproportionately large increase in human infections (compared to the baseline model where 4 hours direct contact is necessary for exposure to occur).

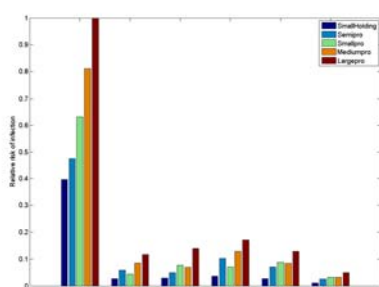


Figure 2: Effect of farm and person type on relative risk of human infection

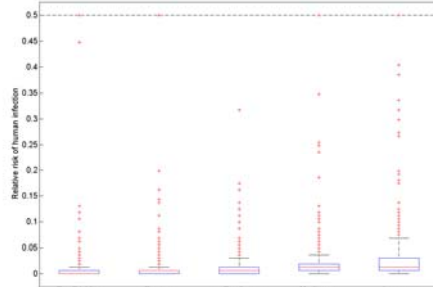


Figure 3: Boxplot to show effect of initial seed farm type on distribution of relative risk of human infection

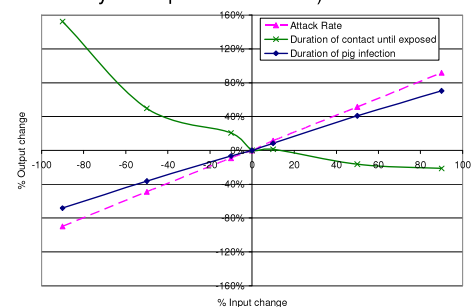


Figure 4: Effect of scenarios on the relative risk of human infection of pH1N1.

## Summary:

The results suggest that while contact with pH1N1 infected pigs on GB farms did not result in any human infections in 54% of iterations, infection can occur. The maximum number of human infections was estimated to be over 150. Large commercial farms have a higher average annual incidence of infection and an outbreak that starts in a large professional farm is likely to lead to more human infections. There is uncertainty associated with the results, due to model assumptions and data gaps, particularly with regards to the threshold for exposure ( $T_{pig}$ ) and attack rate. The scenario analysis highlighted that there are many people who have a short duration of direct contact with pigs (i.e.  $\leq 2$  hours per week), below the threshold for exposure to infection in the baseline model and hence a virus strain that required shorter threshold contact time (i.e.  $< 2$  hours) could result in a great increase in the number of human infections. Reductions in infection could be achieved through focussing on reducing the time infection is present amongst the pig population on the farm.

# A Transport & Lairage model for Salmonella transmission between pigs, applicable to EU member states

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## Introduction

Transport and lairage (T&L) are thought to be important stages for salmonella transmission in the pig production chain. It has been reported that there are significant increases in the prevalence of pigs infected with salmonella between the farm and the slaughterhouse and that pigs can become infected when exposed for less time than the duration of T&L (Berends et al. 1996). There has been little development of the T&L stages in previous pig salmonella QMRAs, mostly relying on simple equations to model a proportional change in infection levels between the farm and slaughterhouse. Here we present a detailed stochastic model for salmonella in pigs during T&L, designed to be able to simulate the effect of interventions. This model is part of a full farm-to-consumption risk assessment for salmonella in pigs, which was requested by the European Food Safety Authority (EFSA) in response to a European Commission mandate. The full model provides an estimate for the risk of human illness from consumption of pork cuts, minced meat and fermented sausages (EFSA, 2010).

## Methods

The T&L model simulates the transmission of salmonella infection within batches of pigs, during transport to the slaughterhouse and subsequent lairage. It uses as an input, a database detailing the number of infected slaughter-age pigs in batches leaving the farm module. The environmental contamination of the trucks and the lairage environment is also simulated, in order to model cross contamination. Each iteration of the model represents one day's worth of pigs going to one slaughterhouse. The model was parameterised using data obtained from MSs, published literature and, where necessary, expert opinion. The computational steps of the model are outlined in the Figure 1.

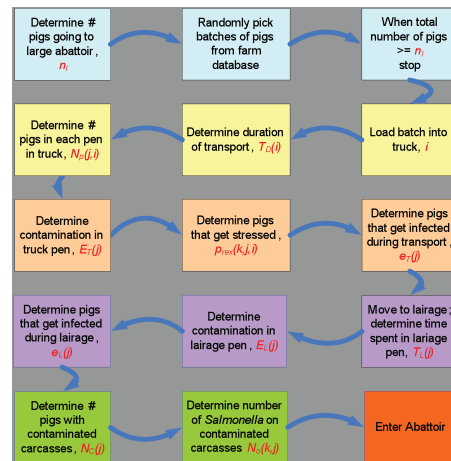


Figure 1: Model overview

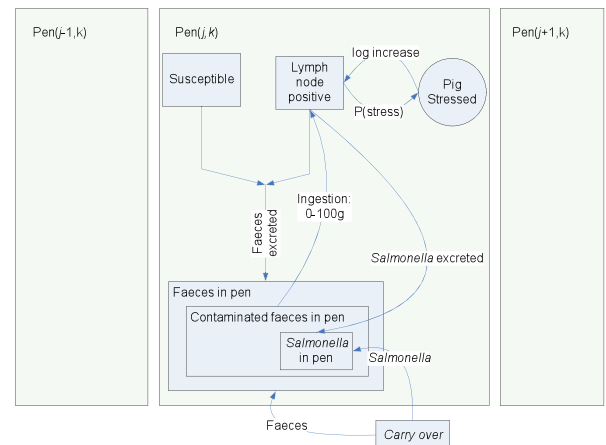


Figure 2: Transmission framework

## Results

Table 1 details the lymph node positive prevalence in batches of pigs at different stages of the T&L model for two case study MSs. Figure 2 shows the distributions of the nonzero increases in lymph node positive batch prevalence during T&L for two case study MSs. Most batches have a small increase (<5%), but a few batches show more than a 70% increase in lymph node positive prevalence. Large changes are more frequent in MS2 than MS1. Figure 3 shows that the change in prevalence during transport for MS2 is inversely proportional to the number of infected pigs at the start of transport. However large increases (>50%) can still be observed with up to 60 initially infected pigs.

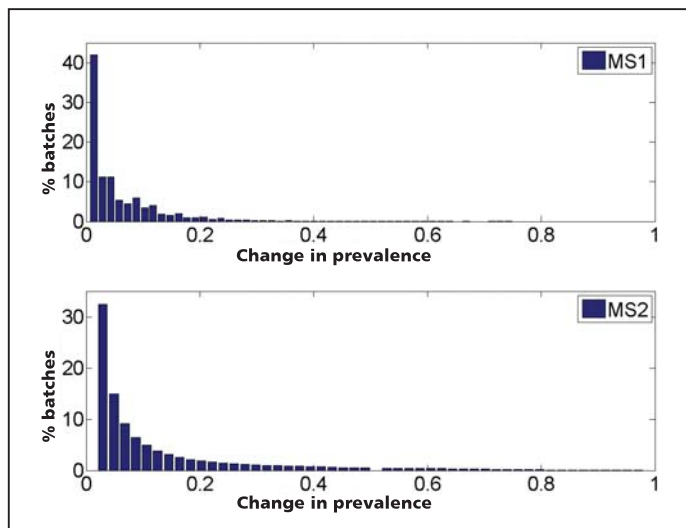


Figure 2: Change in within-batch prevalence during T&L

Table 1: Prevalence of infection

Member state	Mean [5th, 95th percentiles] of prevalence (%)		
	Before transport	After transport	After lairage
MS1	0.43 [0.08, 1.03]	0.62 [0.12, 1.38]	1 [0.2, 2.7]
MS2	16.5 [3.1, 29]	17.6 [4.1, 30.2]	20 [4.9, 35.4]

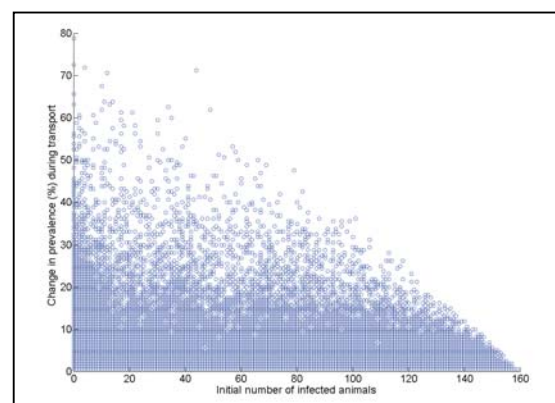


Figure 3: Correlation between change in prevalence and initial number of infected pigs for MS2

## Summary

The T&L model shows that transmission of salmonella can occur during both Transport and Lairage. The estimated increase in batch prevalence between farm and entry to the slaughterhouse, brings the average member state lymph-node prevalence at slaughter in line with the EFSA baseline study results (EFSA, 2008). The change in prevalence is also consistent with Berends et al. (1996) where trials suggest a 0-20% increase in prevalence.

Sensitivity analysis suggests that stress during transport is the most important factor in the model. The main data gap was in estimating carcass contamination, which may have impacted on human health.

Analysis of the full QMRA (EFSA, 2010) suggests that, while T&L interventions show some effect on the prevalence at the start of slaughter, they have little effect on the number of human cases.

## Acknowledgements

We would like to thank our colleagues in the EFSA salmonella in pigs QMRA consortium and also EFSA, Defra, the FSA and the Dutch and Danish governments for funding this work.

## References

Berends et al. (2006). International Journal of Food Microbiology 30: 37-53. EFSA (2010), EFSA QMRA, <http://www.efsa.europa.eu/en/scdocs/scdoc/46e.htm>. EFSA (2008), EFSA baseline survey of Salmonella in slaughter pigs, in the EU, 2006-2007. [http://www.efsa.europa.eu/EFSA/efsa\\_locale-1178620753812\\_1178713190037.htm](http://www.efsa.europa.eu/EFSA/efsa_locale-1178620753812_1178713190037.htm)



# Estimating the per-contact probability of infection by highly pathogenic avian influenza virus during the 2003 epidemic in The Netherlands

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## Introduction

Estimates of the per-contact probability of transmission of Highly Pathogenic Avian Influenza (HPAI) virus between flocks are important for the design of better control and biosecurity strategies.

### Objective

To estimate the probability of a farm being infected with HPAI virus through a given contact from an infectious farm.

## Materials and Methods

We use the data on traced farm contacts (that were not necessarily part of the outbreak control) collected during the epidemic. For each contacted farm, we extracted all its potentially infectious contacts.

- Each farm was characterized by a binary infection status (i.e., response variable)
- A generalized linear model with a binomial distribution and the explanatory variables (contacts) linked by a cloglog function is fitted.

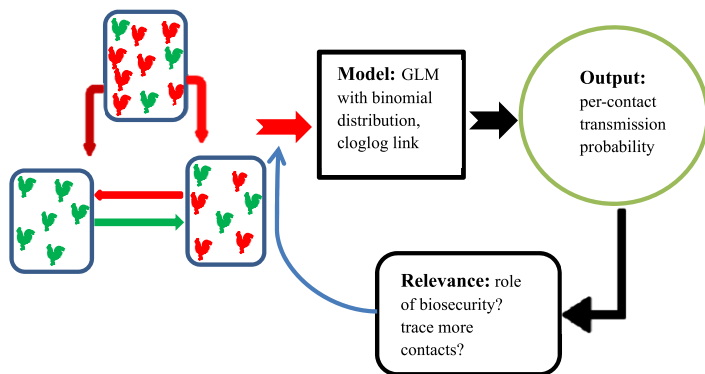


Fig. 1. Schematic view of the approach of this study

## Results

The estimated probabilities (with their accompanying 95% CI) are presented in Table1 and the distribution of (merged) contact-distances is presented in Fig. 2.

Table1: The estimated per-contact transmission probabilities (95% CI)

Variable	Total no. of contacts (no. successful)	Per-contact transmission probability (95% CI)	P-value
Feed contact	267 (101)	0.444 (0.387 - 0.499)	0.00
Egg contact	81 (69)	0.761 (0.681 - 0.840)	0.00
Rendering contact	40 (36)	0.776 (0.646 - 0.890)	0.05
Material and manure contact	13 (11)	0.813 (0.565 - 0.965)	0.13
Advisor and dealer contact	15 (12)	0.738 (0.494 - 0.919)	0.35
Person and family contact	17 (12)	0.739 (0.499 - 0.915)	0.37

\*AIC: 351.78, Residual deviance: 339.78 on 267 degrees of freedom

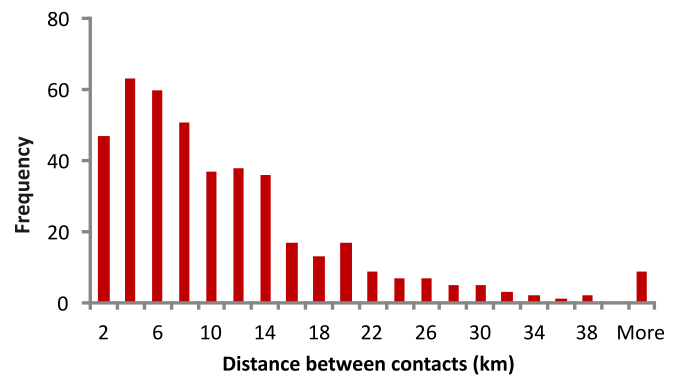


Fig. 2. The distribution of merged infectious contacts

## Discussion and conclusions

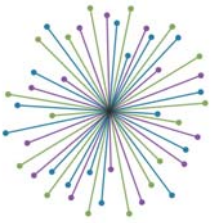
At least one potentially infectious contact was determined for 128 (out of 241) infected and 145 uninfected farms. There is a need to better understand the possible mechanisms of untraced transmission.

The per-contact probabilities obtained here are higher than 0.037 i.e., the upper estimate of the per person-visit transmission probability for the contacts that were part of the outbreak control (te beest et al., ZPH, 2010). These contacts were subjected to thorough biosecurity hence the low estimate.

- This discrepancy provides a scientific support to improve biosecurity measures linked to the contacts

**Relevance:** These estimates can be used to inform studies, such as epidemiological models, that evaluate the impact of improved biosecurity and minimized contact-frequency in controlling HPAI virus spread.

**Next step:** To estimate the per-farm distance-dependent probability of infection by the merged contacts.



CENTRE FOR EVIDENCE-BASED  
VETERINARY MEDICINE  
Putting research into practice

# Characterising the un-owned pet population in the UK

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## Background

Currently the number of dogs and cats in the UK is unknown, although estimates suggest the number of owned dogs and cats to be in the region of 8-10 million for each species (Murray et al., 2010; PFMA, 2010). However, the extent of the un-owned population, comprising strays, ferals and those in shelters and foster care, is currently unknown.

The relationship between the owned and un-owned population is dynamic, with strays and shelter animals being adopted into households, and pet animals being relinquished or abandoned. Un-owned dogs and cats can exist in a variety of environments (Figure 1). These animals may have an increased risk of disease due to stress, poor nutrition, lack of veterinary care and, when housed in shelters, proximity to other animals. It has been suggested that domestic animals may be important in the spread of diseases such as rabies if introduced to the British Isles, and un-owned animals could provide an important reservoir and route of transmission in such circumstances.

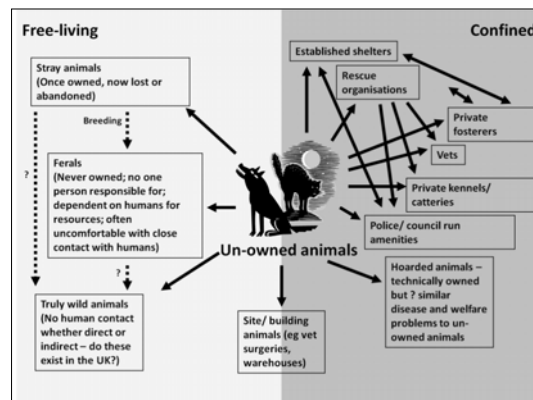
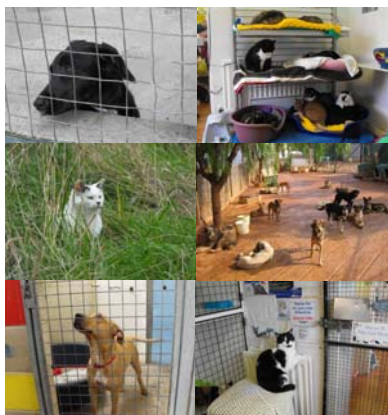


Figure 1: summary of different un-owned dog and cat populations in the UK

## Aim

The study aim is to estimate the number of un-owned dogs and cats in the UK. A secondary aim is to foster a working relationship between the research group and the 'shelter' community, in order to promote the possibility of future research.

## Materials and Methods

### 1. Sampling frame

- England and Wales Charity Commission
- Scottish Charity Commission
- Catchat
- The Dog Rescue Pages

### 2. Questionnaire

- Designed using automated reading software (Cardiff Teleform)
- Piloted with a variety of shelter professionals and volunteers (n=7)
- Distributed by post for 2025 organisations for which postal addresses were available
- Telephone calls and emails to 561 others to obtain postal address

### 3. Compliance

- Visits to shelters and umbrella organisations to publicise project
- Articles in Veterinary Times, Veterinary Record, Veterinary Business Journal
- Website with downloadable PDF of questionnaire and prepaid return envelope
- Facebook profile for project, with links to main project website
- Incentives included with questionnaire (chocolate and pen)

Database (~2500 organisations)  
Rescue, shelter, fostering, sanctuary and care of ferals/ strays



Snowball sampling  
(links from identified sites)



## Conclusion

Database of UK cat and dog 'shelters' compiled using available sampling frames and a modified snowball sampling technique

This will be used to carry out a census of the un-owned cat and dog population in the UK

Both the methodology used and data collection to be a springboard for further work involving this important population of animals

## Acknowledgements:

Thanks very much to The University of Nottingham and Novartis Animal Health for sponsoring this research

## References

- Murray, J. K., Browne, W. J., Roberts, M. A., Whitmarsh, A., Gruffydd-Jones, T. J. (2010). "Number and ownership profiles of cats and dogs in the UK." *Veterinary Record* 166(6): 163-168  
Pet Food Manufacturers Association Annual Report (2010) [http://www.pfma.org.uk/about\\_us/index.cfm?id=65](http://www.pfma.org.uk/about_us/index.cfm?id=65) Accessed 18.01.2010

# Epidemiological consequences of bluetongue serotype 1 incursions to Great Britain

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<sup>1</sup>Institute for Animal Health, Pirbright, UK, <sup>2</sup>The Met Office, Exeter, U.K.; \*email: tom.sumner@bbsrc.ac.uk



## 1. Introduction

The spread of bluetongue serotype 1 (BTV-1) into northern France in 2008-9 raised the risk of an introduction of the serotype to Great Britain (GB). Vaccination has been shown to be an effective method for protecting livestock against BTV and for reducing incidence of disease. However, while an inactivated vaccine against BTV-1 is available, its use in a pre-emptive manner in GB would require the declaration of a restricted or blue zone for the serotype with the associated costs for the livestock industry. Using mathematical models we assessed the risk to GB livestock in the event of an introduction of BTV-1 and evaluated strategies for the deployment of vaccine.

## 2. Modelling Approach

- A stochastic, spatially-explicit model was used to describe the spread of BTV-1 within and between farms in GB [1,2].
- The within-farm model included two host species (cattle and sheep) and one vector population (*Culicoides*).
- Between-farm transmission was described by a Gaussian transmission kernel.
- Vaccination was assumed to reduce the vector-host and host-vector transmission probabilities with 100% efficacy [2].

## 3. Incursion Scenarios

- The average frequency of incursions to each southern county of England over the period May-Oct, 2006-2008 were assessed using a modified version of the Met Office's dispersion model, NAME [3].
- Coastal areas of north-west France were assumed to be source areas of windborne BTV-1 infected *Culicoides* biting midges.
- All counties on the south coast of England were at risk, with the western-most counties (Kent and Sussex) at greatest risk and Cornwall at the lowest risk.
- Simulations were then carried out for three incursion dates (1<sup>st</sup> May, 1<sup>st</sup> July, 1<sup>st</sup> Sept).
- For each date the relative incursion frequencies are used to select the county in which the introduction occurred.

## 4. Vaccination Scenarios

- Thirteen vaccination scenarios were considered
- Baseline scenario: no vaccination
- Six reactive scenarios: uptake 80% or 95% within 20, 50 or 100 km radius of infected farms
- Six pre-emptive scenarios: uptake 50%, 80% or 95% in blue zone BZ 1 or BZ 2, plus reactive vaccination at 95% uptake in a 20km ring

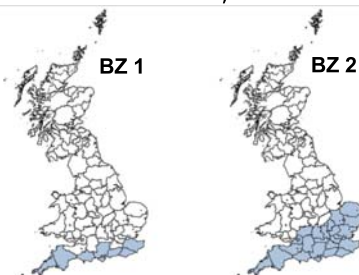


Figure 1. Blue zones (BZ) for BTV-1

## 5. Results

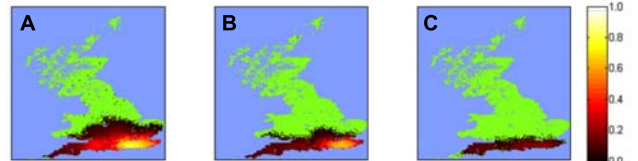


Figure 2. Cumulative risk of spread of BTV-1 in GB in the absence of vaccination for an incursion on A) 1<sup>st</sup> May, B) 1<sup>st</sup> July, C) 1<sup>st</sup> September

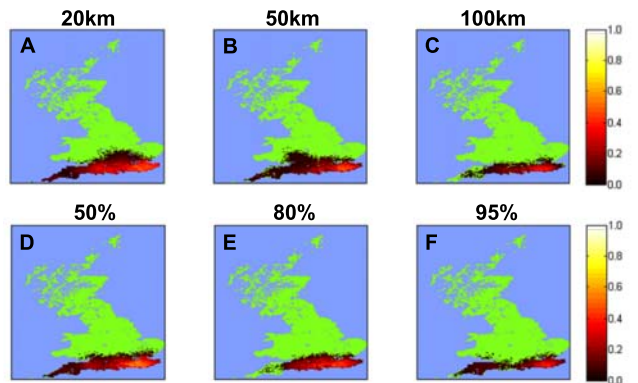


Figure 3. Cumulative risk of spread of BTV-1 in GB for (A-C) reactive scenarios with uptake of 95% (D-F) pre-emptive scenarios using BZ 2, when incursions occur on 1<sup>st</sup> May

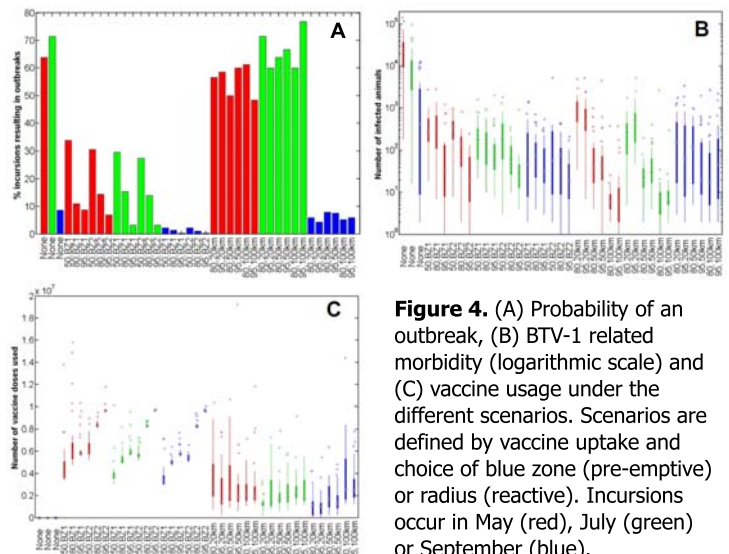


Figure 4. (A) Probability of an outbreak, (B) BTV-1 related morbidity (logarithmic scale) and (C) vaccine usage under the different scenarios. Scenarios are defined by vaccine uptake and choice of blue zone (pre-emptive) or radius (reactive). Incursions occur in May (red), July (green) or September (blue).

## 6. Conclusions

- In the absence of vaccination GB livestock are at risk from an incursion of BTV-1 to the south coast of England.
- The date of incursion significantly affects both the probability that an outbreak will take off and the size of an outbreak.
- The probability of an outbreak is significantly reduced by pre-emptive vaccination but, not by reactive vaccination.
- Both pre-emptive and reactive vaccination reduce outbreak size. The greatest reductions are seen for large-scale reactive vaccination.
- The size of the blue zone does not significantly affect the probability of an outbreak or the number of animals affected.
- The impact of reactive vaccination depends on the timing of the incursion and is greatly reduced for later incursions.



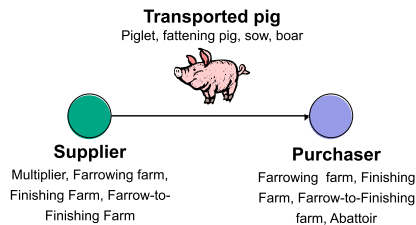
# Infectious disease spread in the pork supply chain

## A modelling approach using network theory

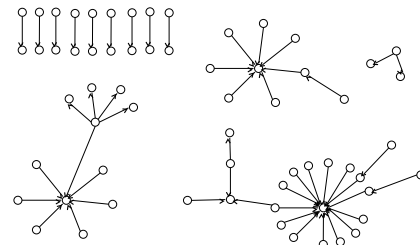
Have the network structure and the initially infected premise an impact on the course of disease spread?

### Data

- 6,892 pig movements between 176 premises



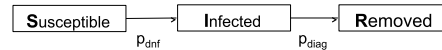
- Recording from 06/2006-05/2009 on weekly basis



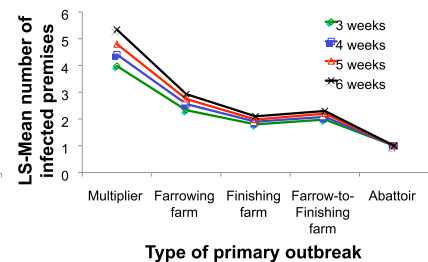
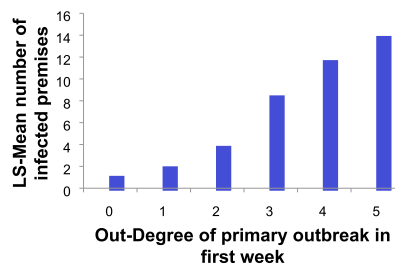
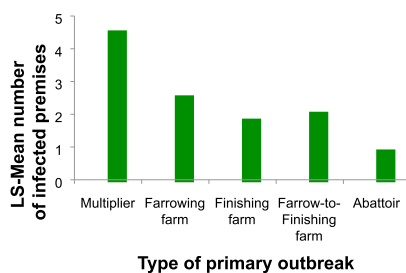
Network structure elements in an exemplarily chosen week

### Method

- SIR-Model, dynamical percolation
- Start at one primary outbreak premise
- Scenarios:
  - Influence of temporal changes in trading relations: Random change in the order of the weeks
  - Randomly chosen primary outbreak
  - Probability of infection ( $p_{inf}$ ): 0, 0.5, 0.6, 0.7, 0.8, 0.9, 1
  - Probability of detection and culling ( $p_{diag}$ ): primary infected premise (3,4,5,6 weeks), secondary infected premise (1,2,3,4 weeks)



### Results



### Conclusion

- General structure of the network constant over time, only slight effect on epidemic.
- Course of the epidemic depends both on type and out-degree of the primary outbreak premise.
- Type of primary outbreak particularly important if long high risk period and high risk of transmission.



# Fitness in horses after carriage driving competitions: individual prediction of fitness to compete.

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<sup>2</sup> current affiliation: GD - Animal Health Service, Netherlands

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I.E.M den Uijl  
GD - Animal Health Service,  
The Netherlands

## Background

Predicting an individual prognosis is common in human medicine, e.g. APGAR score, breast cancer risk or scores for cardiovascular events. While veterinary medicine commonly deals with herds, in some cases individual predictions could be useful. Drivers from carriage driving competitions objected to the invasiveness of measuring rectal temperature (RT). RT is measured as proxy for fitness to compete. A carriage driving competition comprises three parts, of which the marathon, the most demanding part, comes second. The regulations state that horses are fit to compete if RT after the finish of the marathon does not exceed 40°C, to prevent denaturation of proteins and subsequent muscle or metabolic problems.

## Aim

This study aims to replace RT measurements by designing a prediction model to predict the RT at the finish of the obstacle course based on non-invasive measurements. This prediction model, predicting the risk of exceeding RT limits potentially dangerous to the animal, could replace the very invasive and disliked RT measurements at the finish, limiting the burden on drivers as well as animals.

## Methods

Data on physiology (heart rate (HR), RT, color, location in the team and horse/pony) and environment (ambient temperature, humidity, difficulty of the course) were available for 1042 records from 278 horses and ponies over 7 competition days (Fig. 1). The multilevel model, adjusting for repeated measurements and equality of teams, was fitted using manual backwards selection, removing predictors for RT>40°C with p<0.15. Calibration of the models was assessed by plotting the predicted probabilities against their observed probabilities. Shrinkage was adopted to improve calibration by using the slope of the calibration plot as shrinkage factor. The discrimination of the models was assessed using the area under the curve (AUC) of a receiver operating curve (ROC). The prediction score was based on the coefficients of the predictors in the model.

## Results:

Predictors used to calculate the score were: ambient temperature, humidity >60%, horse (as opposed to pony), harnessed at the back, and HR all measured at the finish (Table 1). After shrinkage of the estimates to avoid overfitting (shrinkage factor=1.4), calibration of the score was good. In addition, the area under the curve of the ROC was 0.95 (Fig. 2), indicating good discrimination as well.

Three score categories were formed (Table 2). Animals with low scores, <110 points, were not at risk for exceeding the maximum RT at the finish. Animals with moderate scores, 110-130 points, were at a 9% risk of exceeding the maximum allowed RT. Finally animals with high scores, >130, were at 47% risk of exceeding the maximum limit. The high risk category contained 79% of animals exceeding the RT limit at the finish. All of these recuperated after extra time to cool and extra rest (30min.) under supervision of a veterinarian. The remaining 21% of horses at risk fall in the moderate category and might also benefit from extra rest.

Score	Risk category	Risk
<110 points	Low	0.0
110-130 points	Medium	0.09
>130 points	High	0.47

Table 2 Risk categories

## Example

At a competition with ambient temperature 15°C and humidity 80%, baseline risk for pony singles, pairs and at the back of a four-in-hand would be (15\*2)+10+11=51 points. For horses baseline risk would be slightly higher at 63 points, while for leading horses in a four-in-hand the risk would be decreased by 11 points. Added to this would be their HR at the finish. Consider a single horse in this competition finishing with a HR of 60 beats/min. Baseline risk was (15\*2)+10+11+12=63 points, added to a HR of 60 would be 123 points; the 'medium risk' category. If this same horse would have finished with a heart rate of 76 beats/min, he would have been in the 'high risk' category and would have extra rest.



Photo: Aart van de kamp competing in the marathon.

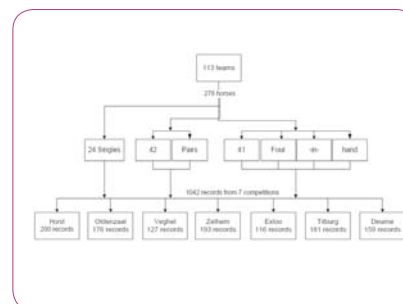


Fig. 1 Hierarchy of the data.

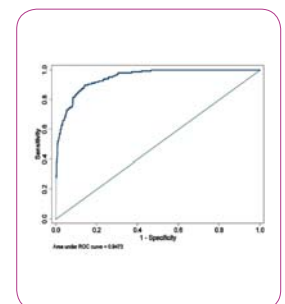


Fig. 2 ROC curve

Parameter	coefficient (95% CI)	Shrunk coefficient	Score
Ambient temperature (°C)	0.24 (0.17 -0.32)	0.2	2
Humidity>60%	1.4 (0.83 -1.9)	1.0	10
Horse	1.6(0.88 -2.4)	1.2	12
Harnessed at the back	1.5(0.67 -2.4)	1.1	11
HR at the finish obstacle course	0.11(0.09 -0.15)	0.1	1

Table 1 Predictors of RT at the finish, estimates and scores.

## Conclusion

This prediction model, predicting the risk of exceeding RT limits potentially dangerous to the animal, could replace the very invasive and disliked RT measurements at the finish, limiting the burden on drivers as well as animals. At the finish only the four risk components need to be measured and the risk score can be calculated. Subsequently, 72% of horses can be cleared, while only 28% will need further rest and examination by a veterinarian. Especially horses at the back of the team, competing in humidity >60% seem to be at high risk of overheating. Officials could advise drivers to cool and provide extra rest for horses located at the back of a team.

# Engaging communities in participatory methods to identify and prioritise issues of concern to owners of working horses in Lesotho

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## Introduction

In 2007, a UK-based equine charity established a training programme in Lesotho covering farriery, saddlery and equine nutrition skills. Lesotho is a small, independent country (population ca. 2.0 million) landlocked within South Africa (Fig 1). It lies entirely more than 1,400m above sea level and is one of the fifty poorest countries in the world. There are an estimated 75,000 horses and ponies, and 144,000 donkeys in Lesotho. **The aim of this study was to use a participatory approach to identify and prioritise the equine issues that were of most concern to owners of working horses in Lesotho.**



Figure 1: Lesotho is an independent country landlocked within the Republic of South Africa

## Objectives

The objectives of the study were to;

- (1) create local horse owner discussion groups;
- (2) create community maps of the areas covered by owner discussion groups;
- (3) facilitate discussion by owners of issues relating to horses;
- (4) enable owners to rank these issues in order of priority so that future educational interventions could be targeted at those topics deemed most important by owners.



Figure 2: Lesotho horse owners were invited to join a participatory discussion

## Methods

Drawing on Participatory Rural Appraisal (PRA) techniques, three one-day participatory workshops for horse owners were organised in three different geographic locations (Mafeteng, Matsieng and Malealea). Owners worked together to create a map of their local area (Fig 4), sketched out on the ground, using locally available materials such as bottle tops, small stones and maize cobs. Features incorporated in the map included roads/tracks, rivers and water sources, location of villages, number of horses in each village and availability of horse related services, such as a farrier, saddler and equine health advice/drugs. Each participant also identified the location of their own home on the constructed map.

As the map was created, owners were encouraged to identify and discuss issues (Figs 2 and 3) associated with owning horses (such as available grazing areas and challenges related to nutrition). These issues were then ranked in order of their relative importance, using a matrix, also drawn on the ground (Fig 5). Issues were written onto cards and then compared against each other in the matrix to identify which was the more important. Owners themselves decided and agreed on the issues and the criteria for their discussion thereby enabling owners to determine those which they perceived as having the greatest impact on horse health and welfare. At the end of the ranking exercise, the number of times each issue had been selected as the priority issue was summed to calculate overall scores for each issue and thereby create a summary ranking scheme.



Figure 3: Lesotho owners gather to discuss issues associated with horses



Figure 4: Lesotho horse owner discussion group creating a local resource map



Figure 5: Lesotho horse owner discussion group ranking priority issues

Table 1: Mafeteng horse owners' issue ranking

Ranking	Issue
1	Mouth problems
2=	Nutrition problems/hunger
	Infectious disease
4=	Foot problems
	Parasites

Table 2: Matsieng horse owners' issue ranking

Ranking	Issue
1	Mouth problems
2	Disease
3	Nutrition problems/hunger
4	Horse husbandry
5	Foot problems

Table 3: Malealea horse owners' issue ranking

Ranking	Issue
1	Mouth problems
2	Nutrition problems/hunger
3	Disease
4	Foot problems
5	Horse husbandry

## Results

In the Mafeteng area, 26 owners attended the discussion group and created a local map showing three villages, with 62 horses. In the Matsieng area, 14 owners attended, creating a local map with ten villages, containing 60 horses. In the Malealea area, where 16 owners attended, a local map incorporating 15 villages covering 163 horses was drawn.

Topics prioritised by each group are shown in tables 1-3. For purposes of aggregation, for each group, issue rankings were converted into scores, taking account of variations in issue definition between groups. Aggregated ranking scores are shown in table 4.

All three discussion groups expressed interest in convening follow up meetings to further explore the issues identified. Participants requested support in the form of learning materials that could help them address the problems and concerns discussed in these initial meetings.

Table 4: Aggregation of results of three participatory owner discussion groups

Category ranked	Discussion group			Total	Rank
	Mafeteng	Matsieng	Malealea		
Mouth	5	5	5	15	1st
Disease (including infectious disease and endo/ectoparasites)	3.5+1.5	4	3	12	2nd
Nutrition	3.5	3	4	10.5	3rd
Feet problems	1.5	1	2	4.5	4th
Husbandry (including tack-associated wounds)	-	2	1	3	5th

## Discussion and conclusions

Whilst there was considerable overlap between groups in the issues identified by owners, priorities did vary somewhat between locations. There is a clear need for owner education on a broad range of topics. Further owner group discussions could refine the scope of topics to be incorporated in learning materials and lead to the establishment of ongoing 'learning communities' whereby horse owners continue to discuss issues and enact locally generated solutions to problems they have identified.



# Prevalence and Risk Factors for Swine Influenza in English Pig Farms

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Pigs are an important reservoir of H1N1, H1N2 and H3N2 influenza viruses, strains of which are endemic in pigs worldwide. Swine influenza virus (SIV) can also contribute to substantial economic loss in pig production due to respiratory disease.

Since 1991, the UK has monitored SIV through a national surveillance programme and identified avian-like H1N1, followed by H1N2, as the predominant strains. The programme is based on passive surveillance and therefore the true proportion of farms in England exposed to the different SIV strains and the prevalence of subclinical infection is difficult to estimate.

In order to plan future surveillance activities, improve preventive and control measures, the epidemiology of SIV in England needs to be better understood.

## Background

**M&M**

In total, 2,787 sera collected from 146 farms were tested for antibodies against avian-like H1N1, H1N2 and human-like H3N2 strains using haemagglutination inhibition (HI) tests. The herd-level case-definition was a reciprocal antibody titre of  $\geq 40$  in at least one growing or finishing pig (i.e. not from sows or weaners) as this indicates recent exposure of the herd to the virus.

Spatial clustering was assessed using the spatial scan statistic.

Risk factor analysis: in the first step, univariable analysis was carried out to identify possible associations between variables and farms status. In the 2<sup>nd</sup> step, variables belonging to the same group relating to farm management or other characteristics were assessed together with multivariable models and the best predicting variables were included in the 3<sup>rd</sup> step, the final multivariable model, which was fitted using backward selection (Mastin et al, 2011).

## Objectives

- To determine the prevalence:
  - of avian-like H1N1, H1N2 and H3N2
  - at herd and animal level
  - and to explore spatial distribution of endemic SIV strains
- To identify risk factors for SIV at herd level



## Results

### 1. Prevalence

In total 52% (CI95%: 46-61%) of farms had evidence of recent exposure or current circulation of H1N1 or H1N2. A total of 19 farms tested positive for both H1N1 and H1N2, and no farm was classified as seropositive for H3N2. Farms in North England were more likely to test positive for avian-like H1N1,  $p < 0.05$ , (Figure 1).

Figure 1: spatial distribution of avian-like H1N1 positive farms

### 2. Herd level risk factors

Through the 1<sup>st</sup> and 2<sup>nd</sup> step of the analysis, the variables listed in Table 2 were found to be associated with the outcome with a p-value  $< 0.1$  (as risk or protective factors), and were included in the multivariable analysis of the 3<sup>rd</sup> step. The final logistic regression model identified an increased likelihood of farm seropositivity for farms sampled in autumn, winter or spring months compared to farms sampled in summer, for farms with more than 18 pigs per water space, and for farms rearing pigs indoors. Decreased likelihood of positivity was found for farms using straw yards (Table 3). There was no evidence of any interaction between variables in the final model.

Table 2: List of potential risk/protective factors for SIV, as identified through univariable and multivariable analysis within variable groups explaining similar characteristics of the farm or of herd management (p-value  $< 0.1$ ):

- Total number of pigs on farm
- Location of sick pen (separate building)
- Number of litters mixed together
- Growers and finishers mixed together
- Stocking density in finishers
- Pigs kept indoors
- Use of straw yards for pigs
- Separation of boars upon entry to the farm
- By-product fed to pigs
- Pigs per feed space (weaners and growers)
- Pigs per water space (growers and finishers)
- Duration of rest from light
- Visitors pig clean
- Years of stockman experience
- Stockman participating in pig events
- Season of farm visit

Table 3: Final multivariable model for SIV risk factors

Variable	Odds Ratio	95% confidence interval	p-value
<b>Pig access to water</b>			
18 finishers or less per water space	1.00	-	-
More than 18 finishers per water space	5.22	1.57 – 17.43	0.01
<b>Season of sampling</b>			
Pigs sampled in the Summer months (July-September)	1.00	-	-
Pigs sampled at other times of the year	2.54	1.09 – 5.95	0.03
<b>Housing type</b>			
No pigs kept indoors	1.00	-	-
At least some pigs kept indoors	3.59	1.11 – 11.57	0.03
No pigs kept in straw yards	1.00	-	-
At least some pigs kept in straw yards	0.30	0.11 – 0.82	0.02

Table 1: Overview of SIV in different age groups

Age group (no of samples)	H1N1	H1N2	at least 1 strain
Weaners (711)	2%	8%	9%
Growers (917)	2%	7%	9%
Finishers (864)	4%	8%	11%
Sows (253)	19%	29%	38%

**References**  
Mastin et al., 2011, PLOS currents influenza, PMID: PMC3032880

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## Discussion

In conclusion, the findings of this study improve our understanding of the current swine influenza situation in England and is the only such epidemiological study in the English pig population in recent years. Compared to estimates made based on passive surveillance, the prevalence found was higher than expected and also the most prevalent strain in the UK was H1N2 and not as expected avian-like H1N1. The high prevalence highlights the importance of subclinical infection, specifically that the

importance of SIV as a significant production disease may have been underestimated in the past.

The findings of the risk factor analysis provide insight into how the spread of SIV can be prevented and will inform future surveillance activities. Given the recent spread of pandemic H1N1 in pigs, the findings will also be of importance to develop control strategies for this novel threat for the pig industry.

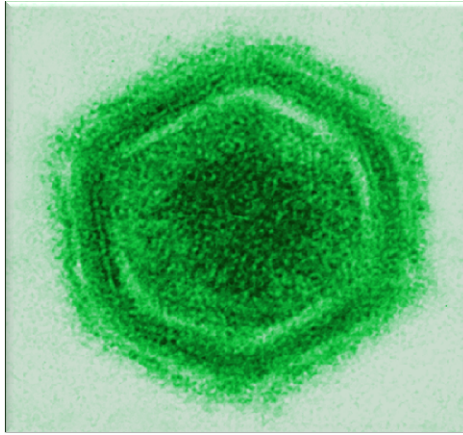
# Proxy driven approach to assess risk of African Swine Fever introduction into EU countries via illegal imports

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Since the outbreaks of African Swine Fever (ASF) in the Caucasus region in 2007 and the subsequent spread to parts of Russia, ASF potentially is a risk for European Union (EU) member states. The most effective transmission of ASF virus is through direct contact between pigs, but the virus can also survive in the environment and for a prolonged time in meat products (reviewed in Costard et al. 2009). Besides legal import of pig and pork products, wild boar, and fomites, illegal import potentially is an important way of introducing ASF into the EU (EFSA ASF working group, 2010; Wieland et al. 2010). However data to estimate the resulting risk objectively following a detailed risk pathway is scarce.

## Background



## M&M

Proxy variables for which data was available in the public domain or could easily be obtained from the respective countries were identified to assess the release and exposure risk for illegal import.

To account for the different weight proxies have on the risk estimate, expert opinion was elicited from experts during a workshop related to the project. Relative weights of proxies were derived through pairwise comparison of proxies.

Quantitative variables were then converted into qualitative risk estimates and then combined using a pre-defined risk matrix. The model structure is implemented in a Microsoft Excel worksheet which is linked to the various tables containing the input data for each country.

To develop a semi-quantitative generic risk assessment framework that allows EU countries to model the risk of introduction via illegal import of pigs and/or pork products

## Objective



**Results** Illegal import of pigs and/or pork products either happens for personal use/consumption or for commercial purpose. For personal use, it was assumed that pork products are the predominant commodity in illegal import. Such imports are linked to tourism or result of residents of at risk countries living and working in the EU wanting to consume products from their home country. Illegal imports for commercial purpose are influenced by the geography of the country, economic factors (price differences of products, demand of products) and the number of residents from countries at risk. The data sources and weights of the different proxies as determined through expert elicitation are presented in Figure 1.

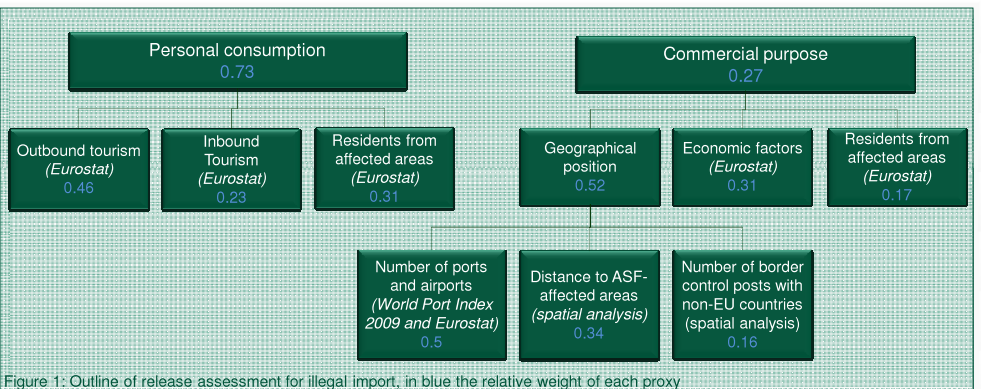


Figure 1: Outline of release assessment for illegal import, in blue the relative weight of each proxy

For the exposure following release of ASF through illegal import, the level of biosecurity in pig production, number of pig workers from ASF affected areas and occurrence of wild boar were considered (Figure 2). The contribution of each proxy to the calculation of the risk of introduction through illegal import are summarised in Figure 3.

Experts considered the release more important than the exposure with weights of 0.66 and 0.34, respectively.

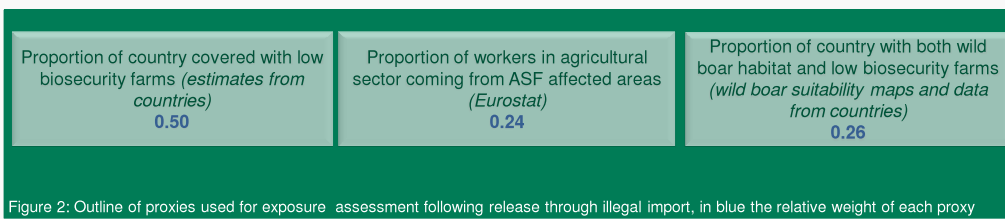


Figure 2: Outline of proxies used for exposure assessment following release through illegal import, in blue the relative weight of each proxy

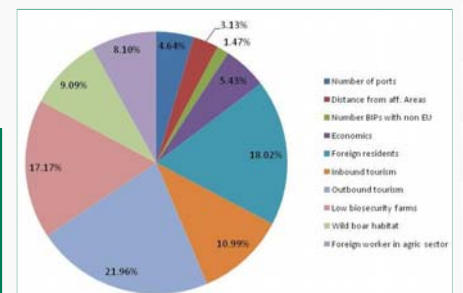


Figure 3: summary of contribution of proxies in release and exposure

## References

Costard et al. (2009), Phil. Trans. R. Soc. B 364, 2683–2696  
EFSA AHAW, Scientific Opinion on African Swine Fever (2010)  
Wieland et al. (2011), PVM 99(1):4-14

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## Discussion & Outlook

The approach presented here will result in relatively crude risk estimates for each country. However, using this standardised approach will allow objective comparison of risk due to illegal import between countries. In the absence of any data on illegal import and trade of pork products, the use of expert elicitation was a good way to gather estimates on the relative importance of proxies identified.

The illegal import pathway presented here is part of an overall generic framework to assess risk of ASF introduction. The other risk pathways, legal import of pigs and pork products, fomites (incl. vehicles and ticks), and wild boar are developed in a similar fashion.

The resulting semi-quantitative models of each pathway are combined in a semi-automated Excel worksheet and will allow EU countries to identify the risk pathways that merit further consideration for risk mitigation. In addition, countries will be able to enter more up to data into the Excel model to replace the open access data that may be out of date.

To validate the approach, for four countries (Spain, UK, Denmark and Poland) the outcomes of the generic risk model will be compared with country specific risk assessments.