# The Centre for Veterinary Epidemiology and Risk Analysis

# The TB Diagnostics and Immunology Research Laboratory

*Biennial Report, 2016-17*

**UCD DUBLIN**

**An Roinn Talmhaíochta, Bia agus Mara**
Department of Agriculture, Food and the Marine

**The Centre for Veterinary Epidemiology and Risk Analysis**

**The TB Diagnostics and Immunology Research Laboratory**

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*S.J. More and D.M. Collins (editors)*

*H.K. Tin (illustrations)*

# Preface

The Department of Agriculture, Food and the Marine (DAFM) provides ongoing financial support to two research units within the UCD School of Veterinary Medicine at University College Dublin:

- The UCD Centre for Veterinary Epidemiology and Risk Analysis (UCD CVERA); and
- The TB Diagnostics and Immunology Research Laboratory

These units each work to support DAFM policy, inspectorate and laboratory staff in the area of animal health. The TB Diagnostics and Immunology Research Laboratory focuses on bovine tuberculosis research. UCD CVERA is a national resource centre, providing policy advice and conducting epidemiological research on a wide range of animal health issues. In addition, UCD CVERA provides general support to government, industry and the veterinary profession (pre- and post-graduation).

This report documents work conducted by, or in association with, these UCD-based research units during 2016 and 2017.

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# Acknowledgements

## *The Centre for Veterinary Epidemiology and Risk Analysis*

UCD CVERA works closely with colleagues from a wide range of organisations, both in Ireland and internationally, and their input is gratefully acknowledged. Staff from each of the following organisations were co-authors, with UCD CVERA staff, of international peer-reviewed scientific papers published during 2016-17:

**University College Dublin**
- UCD School of Veterinary Medicine
- UCD Lyons Research Farm
- UCD School of Agriculture and Food Science
- UCD School of Architecture, Planning and Environmental Policy
- UCD School of Computer Science

**Rest of the Republic of Ireland**
- DAFM State Veterinary Service
- DAFM Veterinary Laboratory Service
- Animal Health Ireland
- Campaign for Responsible Rodenticide Use Ireland CLG
- Ecolab Ireland
- Emel Consulting
- Farmlab Diagnostics
- Forenaghts Stud
- Irish Cattle Breeding Federation
- Marine Harvest Ireland
- Marine Institute
- Population Health Sciences, Royal College of Surgeons in Ireland
- Rentokil Initial Ltd.
- TCD School of Natural Sciences
- TCD Smurfit Institute of Genetics
- Teagasc
- UCC School of Biological, Earth and Environmental Sciences

**United Kingdom**
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- Animal and Plant Health Agency (APHA), United Kingdom
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**Rest of the World**
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- BioEpAR (Biology, epidemiology, and risk analysis in animal health), Nantes, France
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- Immune Solutions Ltd, University of Otago, Dunedin, New Zealand
- Inserm, Paris, France
- Kansas State University College of Veterinary Medicine, Manhattan, Kansas, USA
- Laboratoire Evolution, Génomes, Comportement, Ecologie, Centre National de la Recherche Scientifique (CNRS) - Université Paris-Sud (UMR 9191), Gif sur Yvette, France
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- South Australian Health and Medical Research Institute, Adelaide, Australia
- UNESCO Child and Family Research Centre, School of Political Science and Sociology, National University of Ireland, Galway
- GD Animal Health, Deventer, The Netherlands

## The TB Diagnostics and Immunology Research Laboratory

### Gamma-interferon (IFN-γ) blood testing and research

Staff from the UCD laboratory acknowledge the help and support of Regional Veterinary Office (RVO) staff in providing samples for the IFN-γ test.

### Badger Vaccine development

Staff working on the Badger Vaccine Project acknowledge the contribution and support of Kevin Kenny and colleagues at the mycobacteriology laboratory (DAFM Veterinary Laboratory Service, Backweston, Celbridge, Co. Kildare, Ireland), and Paddy Sleeman of University College Cork for fieldcraft. Mark Chambers, Sandrine Lesellier and staff at the Animal and Plant Health Association (APHA, Weybridge, England) are also thanked for contributing technical expertise for the research programme.

## Personnel

### *The UCD Centre for Veterinary Epidemiology and Risk Analysis*

**Board of management**

Margaret Fitzgerald (HSE) (Chair)
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### *TB Diagnostics and Immunology Research Laboratory*

**Gamma-interferon (IFN-γ) blood testing and research**

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# Overview

## *The Centre for Veterinary Epidemiology and Risk Analysis*

The UCD Centre for Veterinary Epidemiology and Risk Analysis (UCD CVERA) is the national resource centre for veterinary epidemiology in Ireland, located within the UCD School of Veterinary Medicine at University College Dublin. The Centre was initially established as the Tuberculosis Investigation Unit, but has since broadened its remit to cover a wide range of international, national and local animal health matters, including:

- Epidemiological support for the control and eradication of regulatory animal diseases, including the national eradication programme for bovine tuberculosis, and for emergency animal disease preparedness and response;

- Work in support of Animal Health Ireland (**www.animalhealthireland.ie**), which is providing a proactive, coordinated and industry-led approach in Ireland to non-regulatory animal health concerns (such as mastitis, bovine viral diarrhoea and Johne's disease); and

- Epidemiological support for a broad range of other animal health and welfare issues relating to animal health surveillance, on-farm investigations, welfare of farmed livestock and horses, health of companion animals and farmed fish, and international collaboration.

UCD CVERA staff work closely with national policy-makers, both in government and industry. Staff also contribute to training in veterinary medicine, both to undergraduates and postgraduates. A broad range of expertise is represented within the Centre, including database development and management, geographic information systems, statistics, veterinary medicine and epidemiology. The Centre is staffed by employees of University College Dublin and of the Department of Agriculture, Food and the Marine (DAFM).

During 2017, a strategic review of CVERA took place in conjunction with the Department of Agriculture, Food and the Marine, the UCD School of Veterinary Medicine, Animal Health Ireland and other stakeholders. As a result of the review, CVERA has developed clearly defined strategic goals, objectives and expected outcomes for the medium term. These are presented in the CVERA Statement of Strategy 2017-20. Three priority areas have been identified for immediate action, including the establishment of an independent management board, the introduction of systems to improve task management within CVERA, and a communications plan.

For more information please visit: **http://www.ucd.ie/cvera/reports/**

*The TB Diagnostics and Immunology Research Laboratory*

The Tuberculosis Diagnostics and Immunology Research Laboratory based at UCD engages in routine IFN-γ testing of blood samples submitted from Regional Veterinary Offices. Arising from the large number of samples tested each year, there is a focus, in collaboration with CVERA, on analysing the data to gauge the performance of the test under Irish conditions and searching for ways to improve the accuracy of testing. The laboratory also liaises with colleagues working in institutions at home and abroad to share knowledge of the test and keep abreast of any new developments that may impact on its performance.

A second key area of work for the laboratory is the badger vaccine programme of research with the objective to develop a vaccine to control tuberculosis in badgers and to break the link of infection to cattle. Over the course of nine studies with captive badgers, lab personnel have demonstrated as proof of principle that vaccination of badgers with BCG by a number of routes, including oral delivery, generates high levels of protective immunity against challenge with *M. bovis.* The laboratory was also responsible for providing scientific direction to the Co. Kilkenny vaccine field trial. They continue to carry out studies with captive population of badgers to address issues relating to licensing of the vaccine as a veterinary medicine. With the recent announcement of a vaccine implementation strategy by the Minister of Agriculture, Food and the Marine the laboratory will provide key scientific support to underpin the delivery of the strategy.

# Further information

In this report, projects are either:

- Complete, which includes those projects where relevant peer-reviewed papers, or equivalent, have been published in 2016/17, or
- Current, which includes the balance of active projects covering the spectrum from an advanced research concept through to final write-up or manuscripts in press.

Manuscript preparation is conducted in accordance with Uniform Requirements for Manuscripts Submitted to Biomedical Journals of the International Committee of Medical Journal Editors (previously the Vancouver Group). For further information, see **www.icmje.org**. Guidelines for the transparent reporting of specific study types (for example, the CONSORT statement for transparent reporting of trials, **www.consort-statement.org**) are followed.

An up-to-date list of all peer-review papers produced by, or in association with, the UCD Centre for Veterinary Epidemiology and Risk Analysis, and/or The TB Diagnostics and Immunology Research Laboratory is available at **www.ucd.ie/cvera**. Follow us on Twitter @ucd_cvera.

*Susan and Paul*

# Paul White

Paul, our friend and colleague, died in autumn 2016. Paul was a member of CVERA since 1995 and continued to work on his research through to his final days. Paul was a unique individual! He had a boundless curiosity about all things scientific. His enquiring mind was kept busy trying to solve problems by throwing elaborate code at large datasets. Not only did the code have to work, it had to be elegant! Paul's dedication and precision was matched by his passion and enthusiasm for his research; traits shared in his family life. Paul was kind. He had a gentle nature, a good sense of humour and was an eternal optimist! We all have fond memories of Paul and sorely miss him. To Paul's wife Susan and their children Rachel, Jonny, Sarah and Bobby, we offer our sympathies and we thank you for sharing Paul with us for his "research days" in CVERA over the many years.

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# Bovine tuberculosis

# Improving surveillance

## *Field-based surveillance*

### Trends and predictors of large tuberculosis episodes in cattle herds in Ireland

Clegg, T.A. et al.
UCD CVERA

Persistence of bovine tuberculosis (bTB) in cattle is an important feature of *M. bovis* infection, presenting either as herd recurrence or local persistence. One risk factor identified as being associated with the risk of recurrence is the severity of a previous bTB episode. Previous studies have concentrated on chronic herds by considering the duration of an episode. To our knowledge there are no other studies that have looked at predictors of large episodes based on the number of infected animals. The aims of this study are to describe trends in the severity of bTB episodes in Ireland in terms of the number of animals affected per herd and to identify predictors that can distinguish between small and large bTB episodes.

### Future risk of Interferon-Gamma positives

Clegg, T.A. et al.
UCD CVERA

The Interferon-Gamma (IFN-γ) assay has higher sensitivity but lower specificity than the single intradermal comparative tuberculin test (SICTT). Since the specificity of the IFN-γ assay is imperfect, false-positive responses will occur. The aim of this study was to look at the future bovine tuberculosis (bTB) status of animals testing positive to the IFN-γ assay. Field data from a study conducted in 2008 and infected herds that have been tested with the IFN-γ assay since 2015 will be used to look at the future bTB status of IFN-γ positive animals.

### Effectiveness of Interferon-Gamma testing at a herd-level

Clegg, T.A. et al.
UCD CVERA

The Interferon-Gamma assay (IFN-γ) is used as an ancillary test alongside the single intradermal comparative tuberculin test (SICTT) in heavily infected herds. The aim of this study will be to determine whether the use of IFN-γ within a herd has reduced the duration of time that a herd is restricted and whether it has decreased the future risk of a further breakdown. In addition, the characteristics of herds with IFN-γ test positive animals in terms of the herd size and the number of SICTT positives will be examined.

## Validation of the Interferon-Gamma assay

Clegg, T.A. et al.

UCD CVERA

The Interferon-Gamma assay (IFN-γ) is mainly applied in heavily infected herds in Ireland to identify infected animals that are missed by the single intradermal comparative tuberculin test (SICTT). The IFN-γ assay has higher sensitivity but lower specificity than SICTT. The cut-off applied in Ireland is one that maximises the sensitivity of the test. Varying the cut-off point used in the IFN-γ assay will be looked at in respect of the bTB lesion status of slaughtered IFN-γ tested animals.

## The temporal and spatial distribution of singleton reactors in Ireland: exploring the spatial variation in specificity of the single intradermal comparative tuberculin test (SICTT)

McGrath, G. et al.

UCD CVERA

There is evidence that non-specific infection or singleton reactors occur as a result of environmental conditions found on farms close to bog, reclaimed wetland and lakes. If this is the case, they should cluster in space and time. We would expect there to be a spatial consistency which should be discernible. The objective of this study is to determine whether spatial/temporal clustering of singleton reactors exists. If there is a discernible effect, areas of persistent high singleton risk could be defined and assigned back to all herds falling within these areas providing veterinary inspectors with an additional layer of information to aid in deciding how to interpret and manage herds with singleton reactors.

## Recurrence at the 6-month check test

Steenbergen, E. et al.

Quantitative Veterinary Epidemiology Group, Wageningen Institute of Animal Sciences, Wageningen University, Wageningen, The Netherlands

The bovine tuberculosis (bTB) eradication programme in Ireland consists of testing every cattle herd at least annually using the Single Intradermal Comparative Tuberculin Test (SICTT). When one or more positive animal(s) is identified, the herd is then 'restricted'. The herd remains restricted until two consecutive negative herd tests are achieved. Following 'de-restriction' the herd is again free to trade, and tested at 6-month intervals for two years and thereafter returns to annual intervals. At the initial 6-month check test, around 10% of herds are positive each year, possibly due to residual infection that was not detected during the previous episode. The aim of this study was to identify and quantify risk factors for failure at the six-month check test. Based on these results, if possible, predictions for herd status at the six-month check test can be made.

## Factory-based surveillance

### Risk factors for cattle presenting with a confirmed bTB lesion at slaughter, from herds with no evidence of within-herd transmission

Clegg, T.A.[1], Good, M.[2], More, S.J.[1]

[1] UCD CVERA, [2] DAFM

There has been a national bovine tuberculosis (bTB) eradication programme (BTBEP) in Ireland for many years. All cattle herds are tested at least annually using the Single Intradermal Comparative Tuberculin Test (SICTT). Further, abattoir surveillance is conducted on all animals at the time of slaughter. In the Irish BTBEP, a substantial number of confirmed bTB lesions are detected in non-reactor animals, to SICTT, from Officially Tuberculosis Free (OTF) herds at slaughter. In this study we investigate risk factors for non-reactor animals from OTF herds presenting with a confirmed bTB lesion at slaughter, but with no evidence of within-herd transmission. A case-control study was conducted, with animal as the unit of interest. The case animals were all SICTT non-reactor animals slaughtered in 2012, with a confirmed bTB lesion identified during routine abattoir surveillance and with no evidence of within-herd transmission. Control animals were selected from all SICTT non-reactor animals slaughtered in 2012 from OTF herds where no bTB lesion was found. Four controls matched by age (±1 year) and location (county) were randomly selected for each case. A conditional logistic regression model was developed for univariable and multivariable analysis. The final multivariable model included: number of movements, herd type, herd-size, inconclusive reactor status at any previous test, abattoir and time spent in a herd restricted for bTB. The odds of being a case increased with the number of times an animal had moved herds. Animals from suckler herds were significantly more likely to be a case compared to those from beef herds. The odds of being a case decreased with herd-size and increased as the time spent in a restricted herd increased. There were three key conclusions from this study. Firstly, the main risk factors for animals presenting with a confirmed bTB lesion at slaughter were: previous bTB exposure history, previous inconclusive reactor result at the SICTT, the number of herd movements and herd type/size. Secondly, there was very limited evidence that these animals could have been detected any earlier. Finally, there is a need to reconsider the importance of abattoir surveillance during the latter stages of an eradication campaign. As herd prevalence declines, an increasing proportion of herd restrictions will be triggered by a single bTB-lesioned animal, with no evidence of within-herd transmission.

### The herd sensitivity of abattoir surveillance for bovine tuberculosis: simulating the effects of current and potentially modified meat inspection procedures in Irish cattle

Willeberg, P. et al.

Department of Diagnostic and Scientific Advice, National Veterinary Institute, Technical University of Denmark, Copenhagen, Denmark

There are proposals to change current meat inspection (CMI) approaches to visual only inspection (VOI). If this were to occur, it could adversely affect the effectiveness of surveillance for bovine tuberculosis (bTB) in EU member states, both for countries with and without official status of bTB freedom (OTF and non-OTF countries, respectively). This study was conducted to estimate the impact of a change from CMI to VOI in abattoirs on herd-level detection sensitivity in Ireland, a non-OTF country, during 2010 to 2012.

# Improving management of high risk herds

## The performance of the interferon gamma assay when used as a diagnostic or quality assurance test in *Mycobacterium bovis* infected herds

Clegg, T.A.[1], Good, M.[2], Doyle, M.[3], Duignan, A.[2], More, S.J.[1], Gormley, E.[3]

[1] UCD CVERA, [2] DAFM, [3] UCD School of Veterinary Medicine

There are two different contexts in the Irish bTB eradication programme in which the interferon-gamma assay (IFN-γ) is applied. Firstly, the IFN-γ assay is applied routinely to high risk cohorts in herds with four or more reactors to the SICTT. The IFN-γ test is then carried out on blood samples submitted to the laboratory within 8 h of collection (diagnostic testing). Secondly, the use of the IFN-γ assay has recently been extended to test SICTT reactors as part of a general quality assurance (QA) scheme to monitor the performance of the SICTT. Blood samples from reactors are tested one day after blood collection (QA testing). In this study, we analysed the relative performance of the SICTT and IFN-γ when used in parallel as an 8 h diagnostic test and as a 24 h QA test on SICTT reactors. A total of 17,725 IFN-γ tests were included in the analysis (11,658 diagnostic tests and 6,067 QA tests). Of the samples submitted for diagnostic testing, the proportion positive to IFN-γ decreased with the severity of interpretation of the SICTT result. Of the standard reactors that were tested with IFN-γ in the QA programme, 92.2% were positive to the IFN-γ test. Among animals that were SICTT –ve/IFN-γ +ve, 18.9% were positive at post-mortem compared to 11.8% of those that were SICTT +ve (standard reactor)/IFN-γ –ve. These results highlight the risk associated with retaining SICTT –ve/IFN-γ +ve animals, and suggest that prompt removal of these animals is necessary to reduce the potential for future transmission.

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# Supporting studies

## Genetics, milk production

## Evaluation of the methodological quality of studies of the performance of diagnostic tests for bovine tuberculosis using QUADAS adapted for the veterinary field

Downs, S.H.[1], More, S.J.[2], Goodchild, A.V.[1], Whelan, A.O.[1, 3], Abernethy, D.A.[4, 5], Broughan, J.M.[1], Cameron, A.[6], Cook, A.J.[1, 7], de la Rua-Domenech, R.[8], Greiner, M.[9], Gunn, J.[1], Nuñez-Garcia, J.[1], Rhodes, S.[1], Rolfe, S.[10], Sharp, M.[1], Upton, P.[1], Watson, E.[1, 11], Welsh, M.[12, 13], Woolliams, J.A.[14], Clifton-Hadley, R.S.[1], Parry, J.E.[1]

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There has been little assessment of the methodological quality of studies measuring the performance (sensitivity and/or specificity) of diagnostic tests for animal diseases. In a systematic review, 190 studies of tests for bovine tuberculosis (bTB) in cattle (published 1934–2009) were assessed by at least one of 18 reviewers using the QUADAS (Quality Assessment of Diagnostic Accuracy Studies) checklist adapted for animal disease tests. VETQUADAS (VQ) included items measuring clarity in reporting (n = 3), internal validity (n = 9) and external validity (n = 2). A similar pattern for compliance was observed in studies of different diagnostic test types. Compliance significantly improved with year of publication for all items measuring clarity in reporting and external validity but only improved in four of the nine items measuring internal validity (p < 0.05). 107 references, of which 83 had performance data eligible for inclusion in a meta-analysis were reviewed by two reviewers. In these references, agreement between reviewers' responses was 71% for compliance, 32% for unsure and 29% for non-compliance. Mean compliance with reporting items was 2, 5.2 for internal validity and 1.5 for external validity. The index test result was described in sufficient detail in 80.1% of studies and was interpreted without knowledge of the reference standard test result in only 33.1%. Loss to follow-up was adequately explained in only 31.1% of studies. The prevalence of deficiencies observed may be due to inadequate reporting but may also reflect lack of attention to methodological issues that could bias the results of diagnostic test performance estimates. QUADAS was a useful tool for assessing and comparing the quality of studies measuring the performance of diagnostic tests but might be improved further by including explicit assessment of population sampling strategy.

# Methodology and preliminary results of a systematic literature review of ante-mortem and post-mortem diagnostic tests for bovine tuberculosis

Downs, S.H.[1], Parry, J.E.[1], Upton, P.A.[1], Broughan, J.M.[1], Goodchild, A.V.[1], Nuñez-Garcia, J.[1], Greiner, M.[2], Abernethy, D.A.[3, 4], Cameron, A.R.[5], Cook, A.J.[1, 6], de la Rua-Domenech, R.[7], Gunn, J.[1], Pritchard, E.[1], Rhodes, S.[1], Rolfe, S.[8], Sharp, M.[1], Vordermeier, H.M.[1], Watson, E.[1, 9], Welsh, M.[10, 11], Whelan, A.O.[1, 12], Woolliams, J.A.[13], More, S.J.[14], Clifton-Hadley, R.S.[1]

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A systematic review was conducted to identify studies with data for statistical meta-analyses of sensitivity (Se) and specificity (Sp) of ante-mortem and post-mortem diagnostic tests for bovine tuberculosis (bTB) in cattle. Members of a working group (WG) developed and tested search criteria and developed a standardised two-stage review process, to identify primary studies with numerator and denominator data for test performance and an agreed range of covariate data. No limits were applied to year, language, region or type of test in initial searches of electronic databases. In stage 1, titles and available abstracts were reviewed. References that complied with stage 1 selection criteria were reviewed in entirety and agreed data were extracted from references that complied with stage 2 selection criteria. At stage 1, 9,782 references were reviewed and 261 (2.6%) passed through to stage 2 where 215 English language references were each randomly allocated to two of 18 WG reviewers and 46 references in other languages were allocated to native speakers. Agreement regarding eligibility between reviewers of the same reference at stage 2 was moderate (Kappa statistic = 0.51) and a resolution procedure was conducted. Only 119 references (published 1934–2009) were identified with eligible performance estimates for one or more of 14 different diagnostic test types; despite a comprehensive search strategy and the global impact of bTB. Searches of electronic databases for diagnostic test performance data were found to be nonspecific with regard to identifying references with diagnostic test Se or Sp data. Guidelines for the content of abstracts to research papers reporting diagnostic test performance are presented. The results of meta-analyses of the sensitivity and specificity of the tests, and of an evaluation of the methodological quality of the source references, are presented in accompanying papers (Nuñez-Garcia et al., 2017; Downs et al., 2017).

# Meta-analyses of the sensitivity and specificity of ante-mortem and post-mortem diagnostic tests for bovine tuberculosis in the UK and Ireland

Nuñez-Garcia, J.[1], Downs, S.H.[1], Parry, J.E.[1], Abernethy, D.A.[2, 3], Broughan, J.M.[1], Cameron, A.R.[4], Cook, A.J.[1, 5], de la Rua-Domenech, R.[6], Goodchild, A.V.[1], Gunn, J.[1], More, S.J.[7], Rhodes, S.[1], Rolfe, S.[8], Sharp, M.[1], Upton, P.A.[1], Vordermeier, H.M.[1], Watson, E.[1, 9], Welsh, M.[10, 11], Whelan, A.O.[1, 12], Woolliams, J.A.[13], Clifton-Hadley, R.S.[1], Greiner, M.[14]

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Bovine tuberculosis (bTB) in cattle is a global health problem and eradication of the disease requires accurate estimates of diagnostic test performance to optimize their efficiency. The objective of this study was, through statistical meta-analyses, to obtain estimates of sensitivity (Se) and specificity (Sp), for 14 different ante-mortem and post-mortem diagnostic tests for bTB in cattle. Using data from a systematic review of the scientific literature (published 1934–2009) diagnostic Se and Sp were estimated using Bayesian logistic regression models adjusting for confounding factors. Random effect terms were used to account for unexplained heterogeneity. Parameters in the models were implemented using Markov Chain Monte Carlo (MCMC), and posterior distributions for the diagnostic parameters with adjustment for covariates (confounding factors) were obtained using the inverse logit function. Estimates for Se and/or Sp of the tuberculin skin tests and the IFN-γ blood test were compared with estimates published 2010–2015. Median Se for the single intradermal comparative cervical tuberculin skin (SICCT) test (standard interpretation) was 0.50 and Bayesian credible intervals (CrI) were wide (95% CrI 0.26, 0.78). Median Sp for the SICCT test was 1.00 (95% CrI 0.99, 1.00). Estimates for the IFN-γ blood test Bovine Purified Protein Derivative (PPD)-Avian PPD and Early Secreted Antigen target 6 and Culture Filtrate Protein 10 (ESAT-6/CFP10) ESAT6/CFP10 were 0.67 (95% CrI 0.49, 0.82) and 0.78 (95% CrI 0.60, 0.90) respectively for Se, and 0.98 (95% CrI 0.96, 0.99) and 0.99 (95% CrI 0.99, 1.00) for Sp. The study provides an overview of the accuracy of a range of contemporary diagnostic tests for bTB in cattle. Better understanding of diagnostic test performance is essential for the design of effective control strategies and their evaluation.

# Antigen-specific IP-10 release is a sensitive biomarker of *Mycobacterium bovis* infection in cattle

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The most widely used ante-mortem diagnostic tests for tuberculosis in cattle are the tuberculin skin test and the interferon-gamma (IFN-γ) release assay, both of which measure cell-mediated immune responses to *Mycobacterium bovis* infection. However, limitations in the performance of these tests results in a failure to identify all infected animals. In attempting to increase the range of diagnostic tests for tuberculosis, measurement of the cytokine IP-10 in antigen-stimulated blood has previously been shown to improve the detection of *M. tuberculosis* and *M. bovis* infection, in humans and African buffaloes *(Syncerus caffer)*, respectively. In the present study, 60 cattle were identified by the single intradermal comparative tuberculin test as tuberculosis reactors (n = 24) or non-reactors (n = 36) and the release of IFN-γ and IP-10 in antigen-stimulated whole blood from these animals was measured using bovine specific ELISAs. There was a strong correlation between IP-10 and IFN-γ production in these samples. Moreover, measurement of the differential release of IP-10 in response to stimulation with *M. bovis* purified protein derivative (PPD) and *M. avium* PPD distinguished between reactor and non-reactor cattle with a sensitivity of 100% (95% CI, 86%–100%) and a specificity of 97% (95% CI, 85%–100%). These results suggest that IP-10 might prove valuable as a diagnostic biomarker of M. bovis infection in cattle.

# A genome-wide association study for genetic susceptibility to *Mycobacterium bovis* infection in dairy cattle identifies a susceptibility QTL on chromosome 23

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Bovine tuberculosis (bTB) infection in cattle is a significant economic concern in many countries, with annual costs to the UK and Irish governments of approximately €190 million and €63 million, respectively, for bTB control. The existence of host additive and non-additive genetic components to bTB susceptibility has been established. Two approaches i.e. single-SNP (single nucleotide polymorphism) regression and a Bayesian method were applied to genome-wide association studies (GWAS) using high-density SNP genotypes (*n* = 597,144 SNPs) from 841 dairy artificial insemination (AI) sires. Deregressed estimated breeding values for bTB susceptibility were used as the quantitative dependent variable. Network analysis was performed using the quantitative trait loci (QTL) that were identified as significant in the single-SNP regression and Bayesian analyses separately. In addition, an identity-by-descent analysis was performed on a subset of the most prolific sires in the dataset that showed contrasting prevalences of bTB infection in daughters. A significant QTL region was identified on BTA23 (P value >1 × 10$^{-5}$, Bayes factor >10) across all analyses. Sires with the minor allele (minor allele frequency = 0.136) for this QTL on BTA23 had estimated breeding values that conferred a greater susceptibility to bTB infection than those that were homozygous for the major allele. Imputation of the regions that flank this QTL on BTA23 to full sequence indicated that the most significant associations were located within

introns of the *FKBP5* gene. A genomic region on BTA23 that is strongly associated with host susceptibility to bTB infection was identified. This region contained FKBP5, a gene involved in the TNFα/NFκ-B signalling pathway, which is a major biological pathway associated with immune response. Although there is no study that validates this region in the literature, our approach represents one of the most powerful studies for the analysis of bTB susceptibility to date.

## IL-10 suppression of IFN-γ responses in tuberculin-stimulated whole blood from *Mycobacterium bovis* infected cattle

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The measurement of bovine interferon-gamma (IFN-γ) forms the basis of a diagnostic test for bovine tuberculosis where *Mycobacterium bovis* sensitised effector T cells produce IFN-γ following *in vitro* stimulation with tuberculin antigens. In cattle infected with *M. bovis* it is also known that the anti-inflammatory IL-10 cytokine can inhibit in vitro production of IFN-γ leading to a reduced response in the IFN-γ diagnostic test. In order to investigate this in greater detail, whole blood samples from tuberculin skin test positive and negative cattle were stimulated with bovine and avian tuberculin antigens and in parallel with a neutralising anti-IL-10 monoclonal antibody. The results showed that IFN-γ protein levels increased when IL-10 activity was suppressed by Anti – IL-10. By using a standard diagnostic interpretation, the elevated levels of IFN-γ were shown to change the level of agreement between the performance of the single intradermal comparative tuberculin test (SICTT) and IFN-γ assay, depending on the tuberculin treatment. A transcriptomic analysis using RT-qPCR investigated the influence of IL-10 activity on expression of a suite of cytokine genes *(IFNG, IL12B, IL10 and CXCL10)* associated with antigen-stimulated production of IFN-γ. The *IFNG* and *IL12B* genes both experienced significant increases in expression in the presence of Anti-IL-10, while the expression of *IL10* and *CXCL10* remained unaffected.

# Improved understanding of ecology and TB epidemiology

## Badger ecology

### A cautionary note on the use of Minimum Number Alive-Derived Trappability Metrics in wildlife programmes, as exemplified by the case of the European Badger *(Meles meles)*

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Minimum Number Alive (MNA) is a widely used index of abundance and trappability (% of population trapped during a capture session) in mark-recapture programmes. MNA is a negatively biased abundance index, and is sensitive to capture probability and the capture session number. This suggests that MNA-trappability will be positively biased, and that the extent of the bias will vary with true trappability and trapping regime. Inappropriate use of the index may therefore lead to over-optimistic estimates of trappability. In this note, we reiterate the dangers of using MNA-trappability for wildlife practitioners. Using closed-population stochastic simulation, we verify that MNA-trappability indeed provides over-optimistic estimates, especially when true trappability is <70% and the number of trapping occasions is small (<7). Using severely biased trappability estimates could have serious consequences for wildlife conservation, management or vaccination studies. We specifically discuss the implications of this finding in relation to European badger *(Meles meles)* control programmes and recommend that future studies, where possible, use multiple metrics of trappability to assess the consistency of results across estimators.

### Factors affecting European badger movement lengths and propensity: evidence of density-dependent effects?

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The movement ecology of wildlife hosts of zoonotic diseases is important when attempting to manage spill-back infection to domestic animals. We studied badger *(Meles meles)* movements, a reservoir species of bovine tuberculosis, using mark–recapture data at a large spatial scale (755 km²) in Ireland. We investigated both intrinsic (sex, age-class, or weight at capture) and extrinsic (territory size, group size or population density) factors that may have affected either movement length or the propensity to move across territorial boundaries. Such information adds to the evidence base when trying to understand how wild populations maintain and spread infection.

## Push and pull factors driving animal movement in a social mammal, the European badger *(Meles meles)*: the effects of behavioural history, sex, group-composition and group-size

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Understanding factors that impact animal movements across spatially structured populations remains a major goal of population ecology, and is particularly important when key parameters vary with differing ecological conditions. Previous work on badgers, a wildlife host of *Mycobacterium bovis*, in Ireland has demonstrated that population density varies significantly across landscapes and this can be associated with changes in movement parameters. In this study, the socio-spatial structure was studied to reveal what push or pull factors may be involved in badger movement histories. This information is vital in attempting to predict the likely impact of interventions that seek to change the size or composition of groups (e.g. targeted culling) for disease management reasons.

## The three seasons: annual cycles in the feeding behaviour of the European badger

Kelly, D. et al.

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Using data from the UK's Randomised Badger Culling Trial (RBCT) and Ireland's Four Area badger control Project (FAP), this study proposes to identify three feeding seasons within the annual lifecycle of the badger suggesting that it is those seasons that are most likely to drive their behaviour towards the consumption of food (be it natural or man-made e.g. oral vaccine baits).



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# Weather influences trapping success for tuberculosis management in European badgers *(Meles meles)*

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European badgers *(Meles meles)* in Ireland and the UK are a reservoir for *Mycobacterium bovis*, the causative agent of bovine tuberculosis (TB). A number of interventions have been evaluated in attempts to control bovine TB within badger populations, and many of which rely on the capture of badgers. One strategy being implemented within Ireland is intramuscular vaccination using Bacillus Calmette-Guérin (BCG), as an alternative to badger culling. The success of vaccination as a disease control strategy depends on the ability to capture badgers and administer vaccines; thus, trapping success is crucial to effectively vaccinate the population (maximize vaccine coverage). A field vaccine trial was conducted in County Kilkenny, Ireland, from 2010–2013. We used data from this trial to evaluate the association between weather (precipitation and temperature data), badger sett characteristics, and badger trapping success. Approximately 10% of capture efforts resulted in a badger capture. Our results indicate that badger captures were the highest in drizzle, rain, and heavy rain weather conditions, and when minimum temperatures ranged from 3–8 °C. Badger captures were the highest at main setts (large burrow systems), and when sett activity scores were high (qualitative classes 4 or 5). Using local precipitation and temperature data in conjunction with observed sett characteristics provides wildlife managers with guidelines to optimize trapping success. Implementing capture operations under optimal conditions should increase the trapping success of badgers and allow for increased delivery of vaccines to manage bovine TB.

## Environmental dust inhalation in the European badger *(Meles meles)*: Systemic distribution of silica-laden macrophages, pathological changes, and association with *Mycobacterium bovis* infection status

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Chronic inhalation of crystalline silica and silicates may lead to severe lung disease in humans, termed silicosis. The disease is an occupational health concern in miners and related professions worldwide. Silicosis is also a strong risk factor for tuberculosis in humans. Due to its subterranean lifestyle, the European badger *(Meles meles)* is continuously exposed to environmental dust, while this species is also susceptible to tuberculosis, caused by *Mycobacterium bovis*. To date, a thorough investigation of mineral dust retention and its possible implication as a risk factor for mycobacterial infection in badgers has not been performed. The aims of this retrospective histological study were (1) to describe the systemic tissue distribution of silica-laden macrophages (SLMs) in badgers; (2) to compare the amount of SLMs in tissues of badgers of differing *M. bovis* infection status, pulmonary SLM burden and age; and (3) to assess whether inflammation was associated with SLMs. We assessed lung, lymph nodes, liver and spleen of 60 wild-caught badgers of known *M. bovis* infection status for the presence of SLMs using polarizing light microscopy. SLMs were consistently present within the lungs and were widely distributed throughout the lymphatic system. No inflammatory reaction to SLMs, as occurs in human silicosis, was observed in any tissue. Distribution and amount of SLMs were similar between *M. bovis* positive and negative badgers, and we were not able to show an association between the amount of SLMs and *M. bovis* infection status. The amount of SLMs within intra- and extrathoracic lymph nodes was positively associated with the amount of pulmonary SLMs, and with age. This is the first report of substantial and systemic tissue retention of mineral dust particles in a mammalian species lacking associated chronic inflammation (i.e. silicosis). We further highlight different pathogenetic mechanisms underlying silicosis and benign SLM accumulations following siliceous dust inhalation.

## Counts of badgers *Meles meles* in their territories in Wicklow

Sleeman, D.P. et al.

UCC School of Biological, Earth and Environmental Sciences

Counts of badgers emerging from setts were attempted in Co. Wicklow, Ireland. The use of counts can be an easy, non-invasive method of estimating population numbers, without some of the disadvantages associated with other methods such as restraint or cage trapping. The results of this study will be discussed with reference to monitoring badgers numbers.

# Infection control strategies

## Gamma-interferon (IFN-γ) blood testing and research

The gamma-interferon (IFN-γ) assay is used as a tool by ERAD to assist in the eradication of bovine tuberculosis from the national herd. In recent years the Sligo Regional Veterinary Laboratory and the Cork Blood Testing Laboratory (BTL) have been carrying out the first (stimulation) stage of the assay, with blood plasmas transported to UCD for the second stage ELISA step.

In the period 2016-2017, approx. 91,000 blood samples were submitted to the laboratory for testing. This represents close to a 100% increase compared with submission numbers for the previous two years. In the coming years, DAFM has proposed to substantially increase the number of IFN-γ tests carried out in Tb infected herds. With this in mind, the Cork Blood Testing Laboratory (BTL) is setting up to conduct all stages of the assay for samples submitted from the Munster region.

The samples submitted for IFN-γ test currently originate mainly from:
- bovine reactor re-test herds, where the IFN-γ test is used to diagnose infected animals that are missed by the SICTT (false negatives).
- SICTT reactor animals where the IFN-γ test is used to confirm the exposure status of SICTT positive animals.

The correlation between positive SICTT result and positive IFN-γ results exceeds 80%, and provides a useful measure of the quality assurance (QA) of the skin test. The IFN-γ testing on skin test reactors is conducted as part of the ERAD Quality Assurance programme.

The laboratory continues to conduct research with a view to improving the performance of the assay under Irish conditions. A number of key studies have been completed and published in the past two years:

### Performance of the tuberculin skin test and interferon gamma assay in infected herds

Along with colleagues in CVERA, we used data generated from routine submissions to the laboratory to investigate specific aspects of the performance of the IFN-γ tests in infected herds. We analysed the relative performance of the SICTT and IFN-γ when used in parallel as an 8 hour diagnostic test and as a 24 hour QA test on SICTT reactors. A total of 27,776 IFN-γ tests were included in the analysis (21,709 diagnostic tests and 6,067 QA tests). Among the highlights, we showed that the proportions of animals positive to the IFN-γ test decreased with the severity of the SICTT reaction irrespective of timing (8h or 24 h) of the first stage of the assay (antigen-stimulation) post-collection of blood samples. We also found that among animals that were SICTT –ve/IFN-γ +ve, 20.6% were positive at *post-mortem* compared to 11.8% of those that were SICTT +ve /IFN-γ -ve. These results highlighted the risk associated with retaining SICTT –ve/IFN-γ +ve animals, and suggested that prompt removal of these animals is necessary to reduce the potential for future transmission.

### The role of IL-10 cytokine in regulation of IFN-γ production in the Bovigam assay

Although IFN-γ is a key inflammatory cytokine involved in the immune response to *M. bovis* infection, its suppression by another key cytokine, IL-10, appears to be central to the progression of tuberculosis in cattle and humans. We set out to gain a more refined understanding of the role that IL-10 suppression may play in the *in vitro* release of IFN-γ following stimulation of bovine blood with tuberculin, and evaluate its impact on the IFN-γ test readings. The activity of IL-10 was indirectly measured by the PPD stimulation of the blood in the presence of an Anti-IL-10 neutralizing monoclonal antibody known to inhibit the *in vitro* activity of IL-10. Consistent with the results of previous studies our experiments showed that suppression of IL-10 activity resulted in elevated levels of IFN-γ following antigen-stimulation of blood from infected animals with PPD-b and PPD-a. The findings of this study indicate that the underlying nature of this association between IFN-γ production and disease progression may be much more complex than previously considered. As infection progresses the IFN-γ levels recorded in the assay are likely to be a consequence of the interplay

between the pro-inflammatory IFN-γ and anti-inflammatory IL-10. Thus, an animal with tuberculous lesions may present with low IFN-γ levels if circulating levels of IL-10 are high in antigen-stimulated cultures.

*Antigen-specific IP-10 release as a biomarker of* **Mycobacterium bovis** *infection in cattle*
We carried out a collaborative study with colleagues in South Africa to identify additional biomarkers of immune activation with a view to improving the performance of diagnostic assays. In humans, a number of alternatives to IFN-γ have been evaluated for the diagnosis of *M. tuberculosis* infection. Of these, interferon gamma-induced protein 10 (IP-10) has proven particularly noteworthy and measurement of the cytokine IP-10 in antigen-stimulated blood has previously been shown to improve the detection of *M. tuberculosis* and *M. bovis* infection in humans and African buffaloes. In this study, cattle were identified by the single intradermal comparative tuberculin test as tuberculosis reactors (n = 24) or non-reactors (n = 36). The release of IP-10 in antigen-stimulated whole blood was measured using a bovine specific polyclonal antibody ELISA. The results showed that IP-10 synthesis was a robust biomarker of *M. bovis* infection when measured as the differential release of IP-10 in response to stimulation with *M. bovis* purified protein derivative (PPD) and *M. avium* PPD. In addition there was a strong correlation between levels of IP-10 and IFN-γ production in the whole blood cultures. The release of IP-10 proved to be a valuable biomarker of *M. bovis* infection in cattle and measurement of this cytokine may improve the diagnostic sensitivity of whole blood stimulation.

*Additional IFN-γ laboratory research*
The laboratory is involved in studies using the IFN-γ assay to estimate the potency of tuberculin in infected cattle. Data from routine submissions is also being analysed with a view to optimising the performance of the IFN-γ test under Irish conditions, and addressing specific policy related questions. The lab is also engaged in active research with groups based at UCD and elsewhere to use advanced transcriptomic tests to better understand the specific immune responses to infection with tuberculosis.

## The badger vaccine development programme

### The Badger Vaccine Field Trial

Results from the vaccine field trial, which commenced in 2009 to test the efficacy of the oral BCG vaccine in free-living badgers were published in early 2017. A report of the trial was also submitted to DAFM.

The vaccine field trial was conducted in Co Kilkenny to investigate the impact of oral vaccination of free-living badgers against natural-transmitted *Mycobacterium bovis* infection. For a period of three years badgers were captured over seven sweeps in three zones and assigned for oral vaccination with a lipid-encapsulated BCG vaccine (Liporale-BCG) or with placebo. Estimates of prevalence and changing tuberculosis incidence were obtained from the measurements of serological immune responses using the Brock (TB) Stat-Pak test, supplemented with post-mortem examination. The effect of oral vaccination in vaccinated badgers was compared relative to non-vaccinated animals under conditions of natural transmission of infection. The trial site was divided into three zones (A, B and C), each with a different level of vaccine / placebo coverage. Badgers captured in the Zone A were administered placebo by the oral route, while all captured badgers in southern Zone C were vaccinated orally with BCG. Badgers captured in the middle zone (B) were randomly assigned a dose of vaccine, or placebo, on a 50:50 basis. During the trial period, the outcome of interest was incident cases of tuberculosis measured as time to seroconversion events using a lateral flow serology test. Among the vaccinated badgers that seroconverted, the time to seroconversion (median = 413 days) was significantly longer compared with non-vaccinated animals (median = 230 days). Survival analysis (modelling time to seroconversion in Zones A and C) revealed that there was a significant difference in the rate of seroconversion between vaccinated and non-vaccinated badgers throughout the trial period. For badgers enrolled during sweeps 1-2, the Vaccine Efficacy (VE) was 36% (95% CI: -62% – 75%). For badgers enrolled at least a year after the beginning of the

trial, the Vaccine Efficacy was 84% (95% CI: 29% – 97%). At the end of the trial period there were significantly fewer animals with clinical tuberculosis among the vaccinated badgers (Zone C) compared with non-vaccinated badgers (Zone A). The results of this vaccine trial have established for the first time that oral vaccination of free-living badgers can reduce the incidence of tuberculosis in badger populations.

### Vaccine development (studies with captive badgers)

In our most recent completed project with captive badgers, we conducted a vaccine safety study to investigate the distribution of BCG in orally vaccinated badgers over a period of twelve weeks. This is one of a series of pre-regulatory studies required to address the safety of oral BCG vaccination in badgers. Complementary studies were carried out by collaborating groups at APHA UK and ANSES / DGAL, Office for Animal Health (France). The preliminary result showed that there was almost no pathology associated with oral vaccination: an important measure of vaccine safety.

### Vaccination of badgers with BCG Sofia strain

Currently the only available vaccine against TB is the BCG vaccine developed from an infectious *M. bovis* strain isolated from a diseased cow between 1908 and 1921. This is the standard vaccine used worldwide to control tuberculosis, particularly in children. In Ireland, we have shown that vaccination of badgers with BCG results in protection against tuberculosis. For historical reasons there are several different strains of BCG used worldwide. Up until two years ago the BCG Danish strain, manufactured in Denmark, was the only commercially licensed BCG vaccine for use in the EU and the vaccine of choice for delivery to badgers in Ireland and UK. However, production of BCG Danish ceased for a number of years due to a commercial takeover and issues relating to product manufacture. As a result, there is a continuing worldwide shortage of BCG Danish strain, and alternative suppliers of vaccine have been approached. DAFM has access to another widely used BCG strain named Sofia. This is a UNICEF approved vaccine strain used widely in humans in the developing world. Blood from animals that have been vaccinated with different BCG strains (including the Danish strain) do not generally respond in the blood test. However, the situation is not known for the BCG Sofia vaccine. Thus, there is a possibility that animals vaccinated with BCG Sofia will respond in the blood test for TB. This would make it difficult to establish if a positive blood test was due to vaccination or infection. We are currently running a study to determine if seroconversion occurs in the blood of badgers vaccinated with BCG vaccine strain Sofia by the intra-muscular route. The benefit of the study is that it will provide new information on the ability to diagnose TB disease in animals vaccinated with BCG Sofia strain. This will allow DAFM and other agencies to make decisions on surveillance and monitoring of vaccinated badger populations. The study is due for completion in 2019.

### Development of novel badger immunodiagnostics

In addition to the vaccination research programme, the UCD laboratory is also contributing to the development and validation of new serological tests for diagnosis of tuberculosis in badgers (in collaboration with APHA, UK). It has already been established that the sensitivity of immunodiagnostic assays improves as the disease severity increases and that this is more pronounced with the serological based assays. In recent years a number of new serological test platforms have been developed but have not been subject to robust validation. We are contributing samples and expertise to joint Ireland - UK studies to validate a number of these tests.

## Oral vaccination of free-living badgers *(Meles meles)* with bacille Calmette Guérin (BCG) vaccine confers protection against tuberculosis

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A field trial was conducted to investigate the impact of oral vaccination of free-living badgers against natural-transmitted *Mycobacterium bovis* infection. For a period of three years badgers were captured over seven sweeps in three zones and assigned for oral vaccination with a lipid-encapsulated BCG vaccine (Liporale-BCG) or with placebo. Badgers enrolled in Zone A were administered placebo while all badgers enrolled in Zone C were vaccinated with BCG. Badgers enrolled in the middle area, Zone B, were randomly assigned 50:50 for treatment with vaccine or placebo. Treatment in each zone remained blinded until the end of the study period. The outcome of interest was incident cases of tuberculosis measured as time to seroconversion events using the BrockTB Stat-Pak lateral flow serology test, supplemented with post-mortem examination. Among the vaccinated badgers that seroconverted, the median time to seroconversion (413 days) was significantly longer (p = 0.04) when compared with non-vaccinated animals (230 days). Survival analysis (modelling time to seroconversion) revealed that there was a significant difference in the rate of seroconversion between vaccinated and non-vaccinated badgers in Zones A and C throughout the trial period (p = 0.015). For badgers enrolled during sweeps 1–2 the Vaccine Efficacy (VE) determined from hazard rate ratios was 36% (95% CI: -62%– 75%). For badgers enrolled in these zones during sweeps 3–6, the VE was 84% (95% CI: 29%– 97%). This indicated that VE increased with the level of vaccine coverage. Post-mortem examination of badgers at the end of the trial also revealed a significant difference in the proportion of animals presenting with *M. bovis* culture confirmed lesions in vaccinated Zone C (9%) compared with non-vaccinated Zone A (26%). These results demonstrate that oral BCG vaccination confers protection to badgers and could be used to reduce incident rates in tuberculosis-infected populations of badgers.

## Pathogenesis of *Mycobacterium bovis* infection: the badger model as a paradigm for understanding tuberculosis in animals

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Tuberculosis in animals is caused principally by infection with *Mycobacterium bovis* and the potential for transmission of infection to humans is often the fundamental driver for surveillance of disease in livestock and wild animals. However, with such a vast array of species susceptible to infection, it is often extremely difficult to gain a detailed understanding of the pathogenesis of infection - a key component of the epidemiology in all affected species. This is important because the development of disease control strategies in animals is determined chiefly by an understanding of the epidemiology of the disease. The most revealing data from which to formulate theories on pathogenesis are that observed in susceptible hosts infected by natural transmission. These data are gathered from detailed studies of the distribution of gross and histological lesions, and the presence and distribution of infection as determined by highly sensitive bacteriology procedures. The information can also be used to establish the baseline for evaluating experimental model systems. The

European badger *(Meles meles)* is one of a very small number of wild animal hosts where detailed knowledge of the pathogenesis of *M. bovis* infection has been generated from observations in natural-infected animals. By drawing parallels from other animal species, an experimental badger infection model has also been established where infection of the lower respiratory tract mimics infection and the disease observed in natural-infected badgers. This has facilitated the development of diagnostic tests and testing of vaccines that have the potential to control the disease in badgers. In this review, we highlight the fundamental principles of how detailed knowledge of pathogenesis can be used to evaluate specific intervention strategies, and how the badger model may be a paradigm for understanding pathogenesis of tuberculosis in any affected wild animal species.

# The effect of oral vaccination with *Mycobacterium bovis* BCG on the development of tuberculosis in captive European badgers *(Meles meles)*

Chambers, M.A.[1], Aldwell, F.[2], Williams, G.A.[1], Palmer, S.[1], Gowtage, S.[1], Ashford, R.[1], Dalley, D.J.[1], Davé, D.[1], Weyer, U.[3], Salguero, F.J.[4, 5], Nunez, A.[4], Nadian, A.K.[1], Crawshaw, T.[6], Corner, L.A.[7], Lesellier, S.[1]

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The European badger *(Meles meles)* is a reservoir host of *Mycobacterium bovis* and responsible for a proportion of the tuberculosis (TB) cases seen in cattle in the United Kingdom and Republic of Ireland. An injectable preparation of the bacillus Calmette-Guérin (BCG) vaccine is licensed for use in badgers in the UK and its use forms part of the bovine TB eradication plans of England and Wales. However, there are practical limitations to the widespread application of an injectable vaccine for badgers and a research priority is the development of an oral vaccine deliverable to badgers in bait. Previous studies reported the successful vaccination of badgers with oral preparations of $10^8$ colony forming units (CFU) of both Pasteur and Danish strains of BCG contained within a lipid matrix composed of triglycerides of fatty acids. Protection against TB in these studies was expressed as a reduction in the number and apparent progression of visible lesions, and reductions in the bacterial load and dissemination of infection. To reduce the cost of an oral vaccine and reduce the potential for environmental contamination with BCG, it is necessary to define the minimal efficacious dose of oral BCG for badgers. The objectives of the two studies reported here were to compare the efficacy of BCG Danish strain in a lipid matrix with unformulated BCG given orally, and to evaluate the efficacy of BCG Danish in a lipid matrix at a 10-fold lower dose than previously evaluated in badgers. In the first study, both BCG unformulated and in a lipid matrix reduced the number and apparent progression of visible lesions and the dissemination of infection from the lung. In the second study, vaccination with BCG in the lipid matrix at a 10-fold lower dose produced a similar outcome, but with greater intra-group variability than seen with the higher dose in the first study. Further research is needed before we are able to recommend a final dose of BCG for oral vaccination of badgers against TB or to know whether oral vaccination of wild badgers with BCG will significantly reduce transmission of the disease.

# Effect of culling and vaccination on bovine tuberculosis infection in a European badger *(Meles meles)* population by spatial simulation modelling

Abdou, M.[1], Frankena, K.[1], O'Keeffe, J.[2], Byrne, A.W.[2, 3]

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[2] UCD CVERA, [3] Agri-Food and Biosciences Institute, Belfast, Co Antrim, Northern Ireland

The control of bovine tuberculosis (bTB) in cattle herds in the Republic of Ireland (ROI) is partially hindered by spill-back infection from wild badgers *(Meles meles)*. The aim of this study was to determine the relative effects of interventions (combinations of culling and/or vaccination) on bTB dynamics in an Irish badger population. A spatial agent-based stochastic simulation model was developed to evaluate the effect of various control strategies for bovine tuberculosis in badgers: single control strategies (culling, selective culling, vaccination, and vaccine baits), and combined strategies (Test vaccinate/cull (TVC)), split area approaches using culling and vaccination, or selective culling and vaccination, and mixed scenarios where culling was conducted for five years and followed by vaccination or by a TVC strategy. The effect of each control strategy was evaluated over a 20-year period. Badger control was simulated in 25%, 50%, and 75% area (limited area strategy) or in the entire area (100%, wide area strategy). For endemic bTB, a culling strategy was successful in eradicating bTB from the population only if applied as an area-wide strategy. However, this was achieved only by risking the extinction of the badger population. Selective culling strategies (selective culling or TVC) mitigated this negative impact on the badger population's viability. Furthermore, both strategies (selective culling and TVC) allowed the badger population to recover gradually, in compensation for the population reduction following the initial use of removal strategies. The model predicted that vaccination can be effective in reducing bTB prevalence in badgers, when used in combination with culling strategies (i.e. TVC or other strategies). If fecundity was reduced below its natural levels (e.g. by using wildlife contraceptives), the effectiveness of vaccination strategies improved. Split-area simulations highlighted that interventions can have indirect effects (e.g. on population size) in non-treatment areas. Our model suggests that mixed control strategies could maintain infection prevalence to a low level for a considerable period even with a growing population. The model supported the hypothesis that culling strategies appeared to be the most effective method for the control of bTB in badgers using parameters, where available, from ROI, either singly or in combination with other strategies. In this model, the success of a vaccination strategy depended partially upon population density and the proportion of the population infected, therefore an initial culling program (to reduce density and/or remove infected badgers) followed by long-term vaccination may be effective in controlling bTB in badgers.

# Quantification of *Mycobacterium bovis* transmission in a badger vaccine field trial

Aznar, I.[1,2], Frankena, K.[2], More, S.J.[1], O'Keeffe, J.[1,2], McGrath, G.[1], de Jong, M.C.M.[3]

[1] UCD CVERA, [2] DAFM, [3] Quantitative Veterinary Epidemiology Group, Wageningen Institute of Animal Sciences, Wageningen University, Wageningen, The Netherlands

In the UK and Ireland, Bacille Calmette-Guérin (BCG) vaccination of badgers has been suggested as one of a number of strategies to control or even eradicate *Mycobacterium bovis* infection in badgers. In this manuscript, we present the results of a badger field trial conducted in Ireland and discuss how the novel trial design and analytical methods allowed the effects of vaccination on protection against infection and, more importantly, on transmission to be estimated. The trial area was divided into three zones North to South (A, B and C) where vaccination coverages of 0, 50 and 100%, respectively, were applied. Badgers were trapped over a 4 year period. Badgers were assigned to either placebo or vaccine treatment, with treatment allocation occurring randomly in zone B. Blood samples were collected at each capture, and serology was performed in these samples using a chemiluminescent multiplex ELISA system (Enfer test). The analysis aimed to compare new infections occurring in non-infected non-vaccinated badgers to those in non-infected vaccinated ones, while accounting for the zone in which the badger was trapped and the infection pressure to which this individual badger was exposed. In total, 440 records on subsequent trappings of individual non-infected badgers were available for analysis. Over the study period, 55 new infections occurred in non-vaccinated (out of 239 = 23.0%) and 40 in vaccinated (out of 201 = 19.9%) badgers. A Generalized Linear Model (GLM) with a cloglog link function was used for analysis. Statistical analysis showed that susceptibility to natural exposure with *M. bovis* was reduced in vaccinated compared to placebo treated badgers: vaccine efficacy for susceptibility, $VE_S$, was 59% (95% CI = 6.5%–82%). However, a complete lack of effect from BCG vaccination on the infectivity of vaccinated badgers was observed, i.e. vaccine efficacy for infectiousness ($VE_I$) was 0%. Further, the basic reproduction ratio as a function of vaccination coverage (p) (i.e. R(p)) was estimated. Given that the prevalence of *M. bovis* infection in badgers in endemic areas in Ireland is approximately 18%, we estimated the reproduction ratio in the unvaccinated population as R(0) = 1.22. Because $VE_S$ was now known, the reproduction ratio for a fully vaccinated population was estimated as R(1) = 0.50. These results imply that with vaccination coverage in badgers exceeding 30%, eradication of *M. bovis* in badgers in Ireland is feasible, provided that the current control measures also remain in place.

# Characterizing transmission of *Mycobacterium bovis* in a multi-host system

Aznar, I. et al.

UCD CVERA & DAFM

In Ireland, there were expectations that BCG vaccination of badgers in the field would help to reduce *M. bovis* transmission not only within the badger population but also between cattle and badgers. Here, the quantitative role of each of the two host species, cattle and badgers, in the transmission of *M. bovis* in Ireland was assessed. Algorithms to estimate the Next Generation Matrix (NGM) and the basic reproduction ratio for the cattle-badger system were developed and all plausible NGMs describing the transmission between the two species were calculated from prevalence data and the number of cattle herds and badgers, collected at the endemic steady state. Furthermore, some answers to potential queries in relation to adding BCG vaccination to the current eradication programme, and some recommendations for policy makers and other stakeholders are presented.

# The national programme
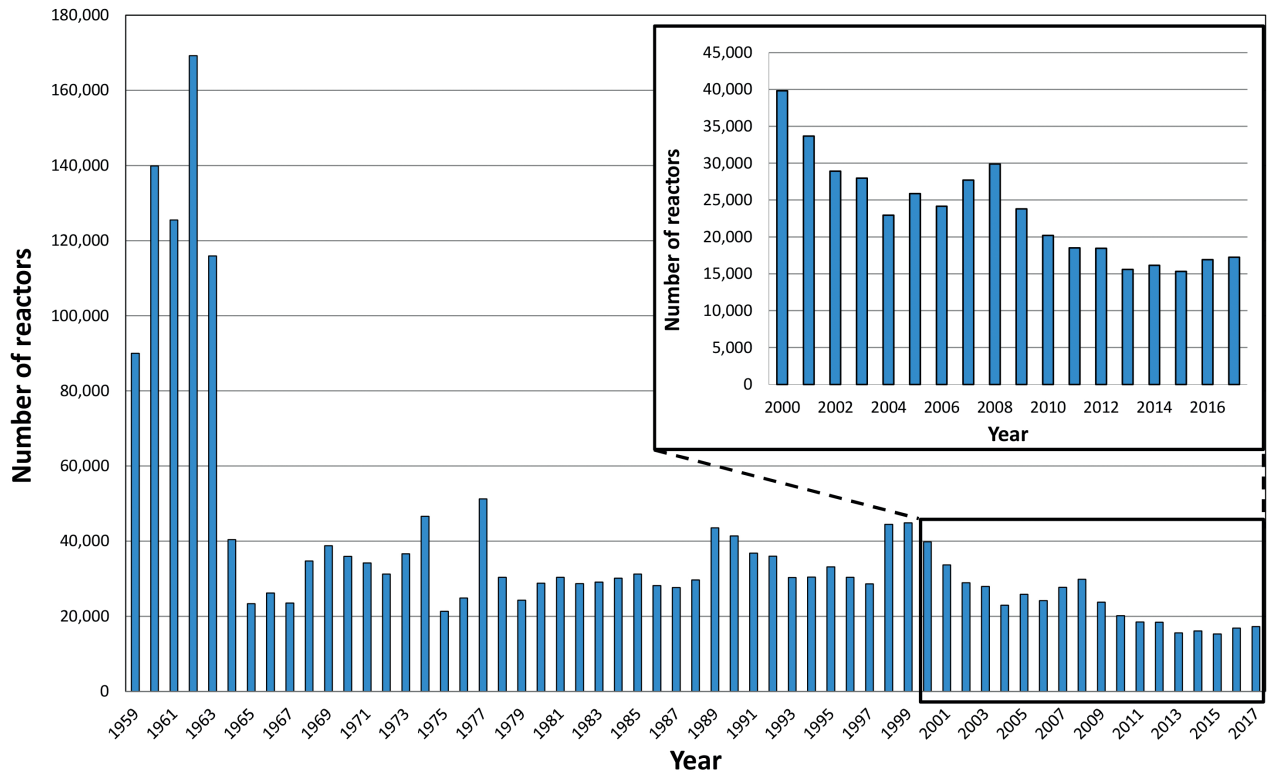


*The number of TB reactors detected in Ireland each year between 1959 and 2017*

## Further evaluation of bovine tuberculosis trends in the UK and the Republic of Ireland, 2003-2015

More, S.J. et al.

UCD CVERA

Bovine tuberculosis (bTB) trends across the UK and the Republic of Ireland have previously been investigated for the period from 1995-2010. Ongoing updates would be useful, providing an evidence base for country-level comparison of bTB trends into the future, especially to describe the effects of the many new policy control initiatives being rolled out across countries and risk areas. This project extends on this earlier work, extending the period to 2015 and also reporting on the frequency, duration and severity of bTB restrictions in each country.

## Investigation of the north Sligo bovine tuberculosis (bTB) outbreak

Doyle, R. et al.

DAFM

Between January and June 2014, six herds in north Co. Sligo were restricted following identification of bovine tuberculosis (bTB). Subsequently, a very significant area-level bTB outbreak developed in the North Sligo area necessitating the need for an enhanced response. This paper describes this bTB outbreak, which occurred in north Co. Sligo during 2014-16, the response undertaken and lessons learned.

# The history of *in vivo* tuberculin testing in bovines: Tuberculosis, a "One Health" issue

Good, M.[1], Bakker, D.[2], Duignan, A.[3], Collins, D.M.[4]

[1] Independent researcher and private consultant, Dun Laoghaire, Co. Dublin & previously affiliated with DAFM, [2] Department of Animal Health, Faculty of Veterinary Medicine, Complutense University of Madrid, Madrid, Spain, [3] DAFM, [4] UCD CVERA

Tuberculosis (TB) is more than 3 million years old thriving in multiple species. Ancestral *Mycobacterium tuberculosis* gave rise to multiple strains including *Mycobacterium bovis* now distributed worldwide with zoonotic transmission happening in both directions between animals and humans. *M. bovis* in milk caused problems with a significant number of deaths in children under 5 years of age due largely to extrapulmonary TB. This risk was effectively mitigated with widespread milk pasteurization during the twentieth century, and fewer young children were lost to TB. Koch developed tuberculin in 1890 and recognizing the possibility of using tuberculin to detect infected animals the first tests were quickly developed. Bovine TB (bTB) control/eradication programmes followed in the late nineteenth century/early twentieth century. Many scientists collaborated and contributed to the development of tuberculin tests, to refining and optimizing the production and standardization of tuberculin and to determining test sensitivity and specificity using various methodologies and injection sites. The WHO, OIE, and EU have set legal standards for tuberculin production, potency assay performance, and intradermal tests for bovines. Now, those using tuberculin tests for bTB control/eradication programmes rarely, see TB as a disease. Notwithstanding the launch of the first-ever roadmap to combat zoonotic TB, many wonder if bTB is actually a problem? Is there a better way of dealing with bTB? Might alternative skin test sites make the test "better" and easier to perform? Are all tuberculins used for testing equally good? Why have alternative "better" tests not been developed? This review was prompted by these types of questions. This article attempts to succinctly summarize the data from the literature from the late nineteenth century to date to show why TB, and zoonotic TB specifically, was and still is important as a "One Health" concern, and that the necessity to reduce the burden of zoonotic TB, to save lives and secure livelihoods is far too important to await the possible future development of novel diagnostic assays for livestock before renewing efforts to eliminate it. Consequently, it is highly probable that the tuberculin skin test will remain the screening test of choice for farmed livestock for the considerable future.

# Further improvements in bovine tuberculosis recurrence in Irish herds

Houtsma, E. et al.

UCD CVERA

During the last few decades in Ireland, there has been a steadily improving situation in the bovine tuberculosis (bTB) eradication programme. However, research has highlighted an ongoing problem of residual infection. Therefore it is of interest to examine how well current measures to prevent recurrence are working. In light of this, the aim of this study is to compare the herd level risk of recurrence of bTB in Ireland between 1998 and 2015.

## Movement in the national herd (analysis and animation)

**McGrath, G. et al.**
UCD CVERA

This study aims to create an animation displaying actual movements of animals within the national herd over a calendar year in Ireland. A previous cohort study looked at movements from County Kerry. This was summarised by the lifetime of the cohort. However, a static image cannot convey the intensity of national movement. An animation showing movements on a daily basis would be a valuable visualisation to emphasise the extent to which herds are connected through the transfer of animals. The animation will distinguish between high risk (farm to farm, farm to mart to farm) and low risk (factory/abattoir) movements. Additional movement statistics highlighting the national spatial and temporal variation of cattle movements within the same period will be published to accompany the animation.

## Spatial structure of farms in Ireland

**McGrath, G. et al.**
UCD CVERA

Most research into transmissible diseases in the national herd makes reference to the fact that farms are often fragmented into multiple disparate parcels, occurring at varying distances away from the 'home farm'. Farms will also have a differing number of neighbours depending on their size and the shape of their perimeters. This study will attempt to analyse and describe the extent of farm fragmentation in Ireland. The number of neighbours and the length of shared boundary with neighbours will also be presented. The metadata will be made available as an online resource accompanying the publication. This will provide a reference point for future epidemiological studies and will provide important metrics for disease modelling.

## Exploring and defining bovine tuberculosis (bTB) clusters in the Republic of Ireland

**McGrath, G. et al.**
UCD CVERA

For many years, bovine tuberculosis (bTB) in cattle in Ireland has been mapped using many techniques from simple point maps and choropleth maps to more complex relative risk surfaces. Clusters are identified visually, but have yet to be given true spatial definitions. This study aims to investigate the characteristics of what constitutes a bTB cluster in terms of size, intensity and duration. This would be a valuable tool in the management of existing clusters and in the early detection of emerging clusters.

## Assessment of the role of collaborative governance in eradicating bovine tuberculosis in Ireland

O'Connor, J. et al.

DAFM

Complex problems in animal health are most effectively solved if stakeholders are willing to together. This work is informed by the theory of collaborative governance which holds that public administrators should directly engage non-state stakeholders in a collective decision-making process that is consensus-oriented, deliberative and that aims to make or implement public policy. Using semi-structured interviews of key stakeholders, we will examine whether collaborative governance can significantly advance efforts to eradicate bovine tuberculosis (bTB) by 2030.

## Cattle movements in Ireland: spatial and temporal patterns

Tratalos, J. et al

UCD CVERA

Movement of animals between herds is one of the primary drivers of the spread of cattle diseases. We examine temporal and spatial patterns of cattle movements in Ireland, identifying temporal trends through the use of time series analysis techniques such as ARIMA modelling. Spatial patterns are analysed through mapping of movements and network modelling. This will provide insights on the movement-mediated mechanism of disease spread in the Irish cattle population and suggest ways in which disease may be better monitored and managed in the future.

## Using network analysis to estimate risk related factors of movement

Tratalos, J. et al

UCD CVERA

Animal movement is one of the drivers of the spread of cattle diseases such as paratuberculosis and bovine tuberculosis (bTB). This study will use a network based approach to examine cattle movements and how network characteristics such as in-degree and out-degree centrality, combined with information about the disease status of individual herds over time, help us to understand the dynamics of disease and to target surveillance appropriately.

# Density of TB incidence



*Legend:*

- < 0.005
- 0.005 - 0.009
- 0.01 - 0.014
- 0.015 - 0.019
- 0.02 - 0.024
- 0.025 - 0.029
- 0.03 - 0.034
- 0.035 - 0.039
- 0.04+
- Non-agricultural land

*Density of TB incidence per square km during 2016 (kernel density with search radius at 10km)*

| | |
|---|---|
| | < 0.005 |
| | 0.005 - 0.009 |
| | 0.01 - 0.014 |
| | 0.015 - 0.019 |
| | 0.02 - 0.024 |
| | 0.025 - 0.029 |
| | 0.03 - 0.034 |
| | 0.035 - 0.039 |
| | 0.04+ |
| | Non-agricultural land |

*Density of TB incidence per square km during 2017 (kernel density with search radius at 10km)*

# APT per DED
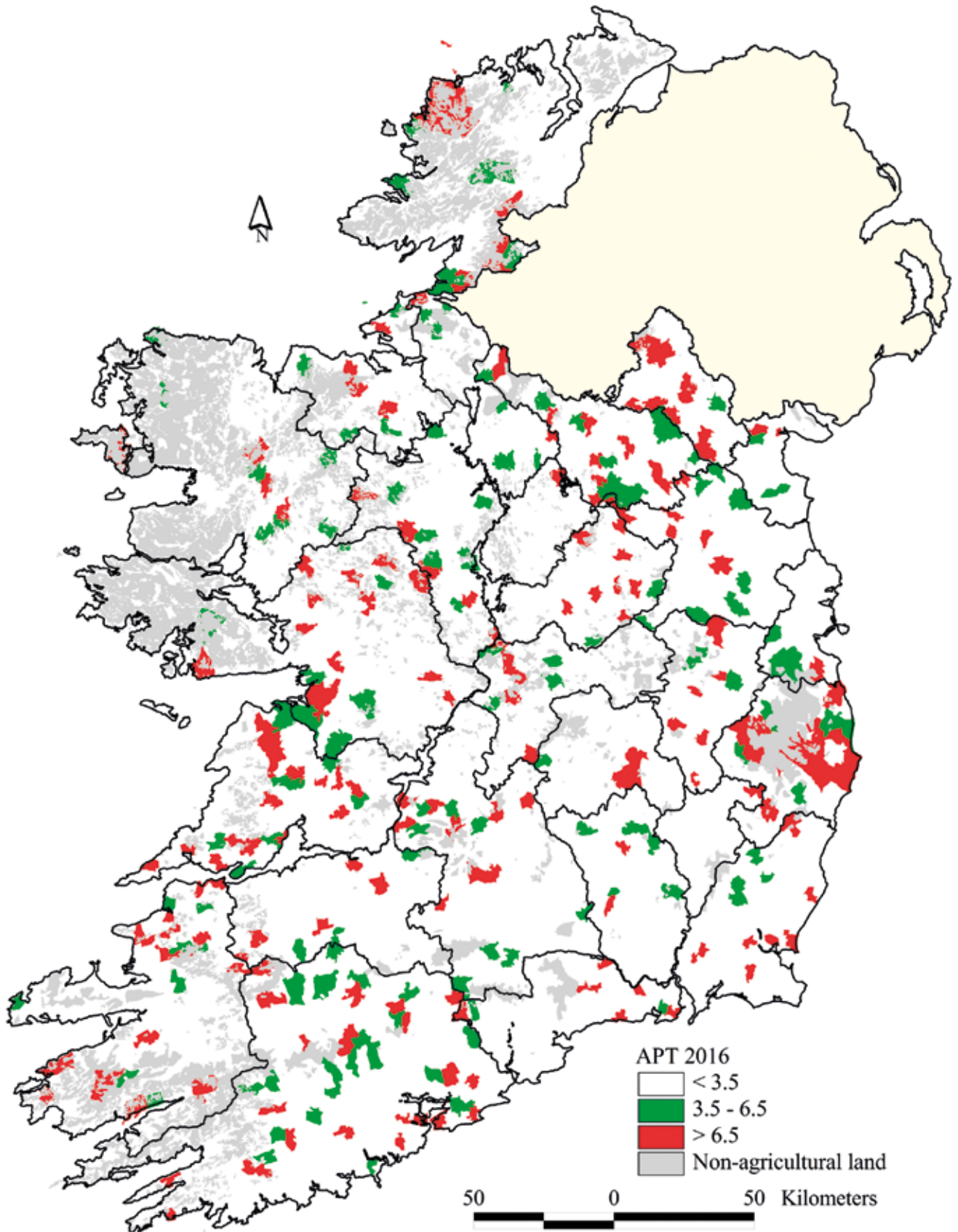


APT 2016

| | |
|---|---|
| ☐ | < 3.5 |
| 🟩 | 3.5 - 6.5 |
| 🟥 | > 6.5 |
| ▦ | Non-agricultural land |

50     0     50   Kilometers

*APT (reactors per 1000 tests) per district electoral division, 2016*

APT 2017

| | |
|---|---|
| ☐ | < 3.5 |
| 🟩 | 3.5 - 6.5 |
| 🟥 | > 6.5 |
| ⬜ | Non-agricultural land |
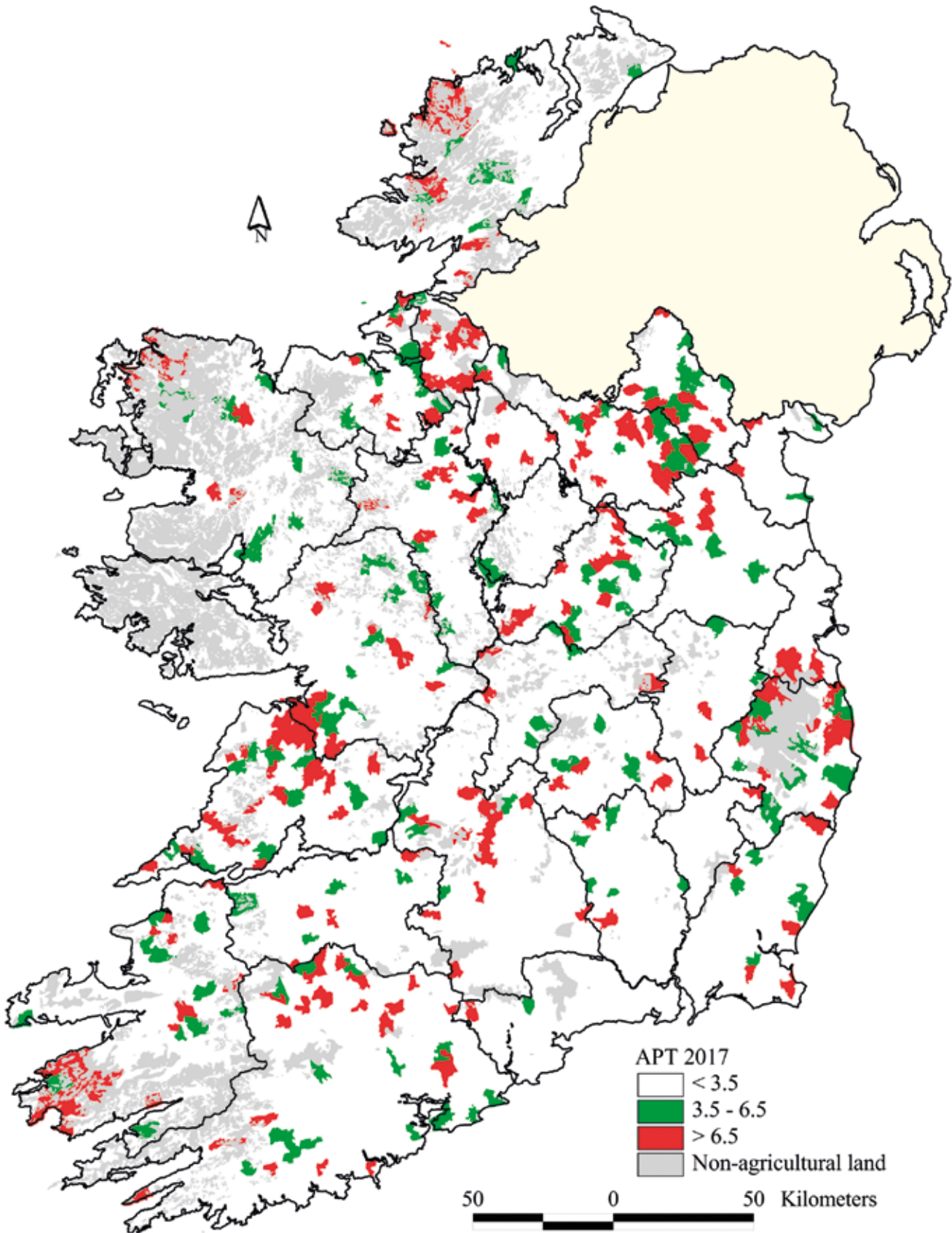
*APT (reactors per 1000 tests) per district electoral division, 2017*

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# Non-regulatory cattle health issues

## Biosecure diseases

# Biosecure diseases

## *Bovine viral diarrhoea (BVD)*

### Prevalence of Bovine Viral Diarrhoea Virus (BVDV), Bovine Herpes Virus 1 (BHV 1), Leptospirosis and Neosporosis, and associated risk factors in 161 Irish beef herds

Barrett, D.[1], Parr, M.[2], Fagan, J.[3], Johnson, A.[3], Tratalos, J.[4], Lively, F.[5], Diskin, M.[2], Kenny, D.[2]

[1] DAFM, [2] Teagasc, [3] DAFM Veterinary Laboratory Service, [4] UCD CVERA, [5] Agri-Food and Biosciences Institute, Belfast, Co. Antrim, Northern Ireland

There are limited data available, in Ireland or elsewhere, to determine the extent of exposure to various endemic diseases among beef cows and factors associated with exposure to causative pathogens. The objectives of this study were to determine the herd and within herd prevalence of Bovine Viral Diarrhoea Virus (BVDV), Bovine Herpes Virus 1 (BHV-1), Leptospirosis and Neosporosis in a large scale study of commercial beef herds on the island of Ireland, and to examine herd level factors associated with exposure to these pathogens in these herds. The average number of cows tested per herd was 35.5 (median 30). Herd level seroprevalence to Bovine Herpesvirus-1(BHV-1), Bovine Viral-Diarrhoea Virus (BVDV), Leptospirosis and Neosporosis was 90%, 100%, 91% and 67%, respectively, while the mean within herd prevalence for the these pathogens was 40%, 77.7%, 65.7% and 5.7%, respectively. The study confirms that the level of seroconversion for the four pathogens of interest increases with herd size. There was also evidence that exposure to one pathogen may increase the risk of exposure to another pathogen. Herd level seroprevalences were in excess of 90% for BVDV, BHV-1 and Leptosporosis. Larger herds were subject to increased exposure to disease pathogens. This study suggests that exposure to several pathogens may be associated with the further exposure to other pathogens.

### Factors associated with the loss of Negative Herd Status (NHS) for BVD in Irish herds during 2017

Barrett, D. et al.

DAFM

Over 70,000 herds in Ireland have achieved Negative Herd Status (NHS) for BVD. NHS is defined as a herd that had no BVD virus positive calves in the previous 12 months and all eligible animals have either a direct or indirect BVD negative status. In 2017, 548 herds with NHS status at the beginning of 2017 lost that status following disclosure of a BVD virus positive calf in those herds. The primary objective of this study is to determine what factors are associated with the loss of BVD NHS.

# Temporal trends in the retention of BVD+ calves and associated animal and herd-level risk factors during the compulsory eradication programme in Ireland

Clegg, T.A.[1], Graham, D.A.[2], O'Sullivan, P.[3], McGrath, G.[1], More, S.J.[1]

[1] UCD CVERA, [2] Animal Health Ireland, [3] Irish Cattle Breeding Federation

The national BVD eradication programme in Ireland started on a voluntary basis in 2012, becoming compulsory in 2013. The programme relies on accurate identification and prompt removal of BVD+ calves. However, a minority of herd owners have chosen to retain BVD+ animals (defined as still being alive more than seven weeks after the date of the initial test), typically with a view to fattening them to obtain some salvage value. During each year of the programme, additional measures have been introduced and implemented to encourage prompt removal of BVD+ animals. The objective of this study was to describe temporal trends in the retention of BVD+ calves and associated animal and herd-level risk factors during the first three years of the compulsory eradication programme in Ireland. The study population included all BVD+ calves born in Ireland in 2013–2015. A parametric survival model was developed to model the time from the initial BVD test until the animal was slaughtered/died on farm or until 31 December 2015 (whichever was earlier). A total of 29,504 BVD+ animals, from 13,917 herds, were included in the study. The proportion of BVD+ animals that were removed from the herd within 7 weeks of the initial test date increased from 43.7% in 2013 to 70.3% in 2015. BVD+ animals born in 2015 had a much lower survival time (median = 33 days) compared to the 2013 birth cohort (median = 62 days), with a year on year reduction in survival of BVD+ calves. In the initial parametric survival models, all interactions with herd type were significant. Therefore, separate models were developed for beef and dairy herds. Overall the results of the survival models were similar, with birth year, BVD+ status, herd size, county of birth and birth month consistently identified as risk factors independent of herd type (beef or dairy) or the numbers of BVD+ animals (single or multiple) in the herd. In addition, the presence of a registered mobile telephone number was identified as a risk factor in all models except for dairy herds with a single BVD+, while the sex of the BVD+ calf was only identified as a risk factor in this model. Significant progress has been made in addressing the issue of retention of BVD+ calves, however, there is a need for further improvement. A number of risk factors associated with retention have been identified suggesting areas where future efforts can be addressed.

## Quantifying the risk of spread of bovine viral diarrhoea virus (BVDv) between contiguous herds in Ireland

Graham, D.A.[1], Clegg, T.A.[2], Thulke, H.-H.[3], O'Sullivan, P.[4], McGrath, G.[2], More, S.J.[2]

[1] Animal Health Ireland, [2] UCD CVERA, [3] Helmholtz Centre for Environmental Research - UFZ, Department of Ecological Modelling, Leipzig, Germany, [4] Irish Cattle Breeding Federation

The control of bovine viral diarrhoea virus (BVDv) mainly focuses on the identification and restriction of persistently infected (PI) animals. However, other transmission pathways can also result in new breakdowns, including the movement of animals pregnant with PI calves (Trojan animals) and the spread of infection between contiguous farms. Contiguous spread is likely an important problem in the BVD eradication programme in Ireland, given the spatial distribution of residual infection, and the highly fragmented nature of land holdings on many Irish farms. In this study, we seek to quantify the risk of BVD spread between contiguous herds in Ireland. Multivariable logistic models were used to estimate the risk of a herd having BVD positive calves in January to June 2014 (the study period) when contiguous to a herd that had at least one BVD positive calf born in 2013. The models included risk factors relating to the study herd and to neighbouring herds. Separate multivariable models were built for each of four "PI-neighbour" factors relating to the presence of BVD+ animals and/or the presence of offspring of PI breeding animals. In total, 58,483 study herds were enrolled. The final model contained the province, the log of the number of calf births born during the study period, the number of cattle purchased between January 2013 and January 2014, and with a two-way interaction between the number of animals of unknown BVD status in the study herd and the PI-neighbour risk factor. When the number of PI-neighbour herds was used as the PI-neighbour risk factor, the odds ratio (OR) associated with the number of PI-neighbour herds ranged from 1.07 to 3.02, depending on the number of unknown animals present. To further explore the risk associated with PI-neighbour factors, the models were repeated using a subset of the study herds (n = 7440) that contained no animals of unknown status. The best fitting model including "any PI-neighbour" as the PI-neighbour factor and also contained the log of the number of calf births born during the study period and the number of cattle purchased. The OR associated with "any PI-neighbour" was 1.92 (95% C.I. 1.37–2.70). This study provides the first quantitative information on the risks posed by the presence of BVD+ animals in neighbouring herds and also highlights the importance of clarifying the BVD status of animals that have not yet been tested in the context of the Irish eradication programme.

## Quantifying the role of Trojan dams in the between-herd spread of bovine viral diarrhoea virus (BVDv) in Ireland

Reardon, F.[1], Graham, D.A.[2], Clegg, T.A.[1], Tratalos, J.A.[1], O'Sullivan, P.[3], More, S.J.[1]

[1] UCD CVERA, [2] Animal Health Ireland, [3] Irish Cattle Breeding Federation

A compulsory national programme to eradicate bovine viral diarrhoea virus (BVDv) began in Ireland on 1 January 2013. The objective of the current study was to quantify the role of Trojan dams (animal(s) not persistently infected (PI) with BVDv but carrying PI foetus(es) and introduced to the herd while pregnant with the PI foetus(es)) in the farm-to-farm spread of BVDv in Ireland, and to identify herd-level risk factors for producing or introducing a Trojan dam. The study population included all BVD+ calves born in Ireland between 1 January 2013 and 31 December 2015, along with their

dams. BVD+ calves included all calves on the national programme database with an initial positive or inconclusive virus test, without a confirmatory re-test (status BVDPOS) and those with an initial positive or inconclusive test and a positive confirmatory test (status BVDPI). The Trojan status of dams was determined after considering their history of movement and of potential BVDV exposure, relative to a defined window of susceptibility (WOS; days 30 to 120 of gestation). During 2013-15, there were 29,422 BVD+ birth events to dams that were not themselves BVD+, including 2,526 (8.6%) most-likely attributable to Trojan dams. The percentage of these birth events attributable to Trojan dams was significantly different (P < 0.001) between years, being 7.1% in 2013, 9.2% in 2014 and 10.6% in 2015. During 2013, in 9.9% of herds with one or more BVD+ birth to non-BVD+ dams, at least one of these births was attributed to a Trojan dam. In 2014 and 2015, the percentages were 11.8% and 13.3%, respectively. In 2013, in 7.8% of herds with one or more BVD+ birth to non-BVD+ dams, all of these births were attributable to Trojan dams. In 2014 and 2015, the percentages were 9.2% and 10.7%, respectively. A logistic GEE regression identified dam parity, herd size and an interaction between herd type and season as significant predictors for the birth of a BVD+ calf to a Trojan dam. Significant predictors for the sale of a Trojan dam from BVD+ herds included those selling more than one pregnant female and those with more than 2 BVD+ animals in the herd. Introduction of pregnant adult females is a potential source of BVD+ births in BVD-free herds and may add to the burden of infection in non-BVD-free herds. Addressing this route of transmission will be critical for herds that are now free of infection and wish to continue to purchase animals without introducing it.

## Epidemiological principles for tackling Bovine Viral Diarrhoea virus (BVDv) at the national level: current international strategies and Irish perspectives

Reardon, F. et al.

UCD CVERA

There are three main objectives in tackling endemic bovine viral diarrhoea (BVD) at the national level, including accurately identifying herds infected with BVD virus (BVDv), resolving infection within these herds by identifying and eliminating persistently infected (PI) animals, and on-going monitoring to ensure that herds remain free from BVDv. This study reviews international experiences in BVD eradication under each of these three objectives, and their potential implications for eradication of BVDv in the Republic of Ireland.

## Potential infection-control benefit of measures to mitigate the risk posed by Trojan dams in the Irish BVD eradication programme

Reardon, F. et al.

UCD CVERA

In the epidemiology of bovine viral diarrhoea (BVD), Trojan dams (animals that are not persistently infected (PI) with BVD virus but carrying PI foetuses) are a vehicle through which infection can be transmitted from farm-to-farm. In the Irish BVD eradication programme, there have thus far been no specific measures taken to address the role of these animals in the spread of infection. The current study was undertaken to assess the effectiveness of movement restrictions on female animals for varying periods following removal of persistently infected (PI) animals from infected herds, in preventing Trojan births in other herds, and the proportionality of such measures in terms of the total number of herds and animals affected by these measures.

## STOC free: An innovative framework to compare probability of freedom from disease in heterogeneous control programmes

Santman, I. et al.

Utrecht University and GD Animal Health, the Netherlands

Several European countries have implemented national or regional surveillance, control, or eradication programmes for endemic infections of cattle. These programmes bring tangible benefits to participating farmers and regional economies, but also create difficulties for intra-community trade, as free trade has the potential to reintroduce infectious agents into regions where freedom has been achieved. This project aims to develop and validate a framework that enables a transparent and standardized comparison of confidence of disease freedom for control programmes across herds, regions or countries.

## Eradicating BVD, reviewing Irish programme data and model predictions to support prospective decision making

Thulke, H.-H.[1], Lange, M.[1], Tratalos, J.A.[2], Clegg, T.A.[2], McGrath, G.[2], O'Grady, L.[3], O'Sullivan, P.[4], Doherty, M.L.[3], Graham, D.A.[5], More, S.J.[2]

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Bovine Viral Diarrhoea is an infectious production disease of major importance in many cattle sectors of the world. The infection is predominantly transmitted by animal contact. Postnatal infections are transient, leading to immunologically protected cattle. However, for a certain window of pregnancy, *in utero* infection of the foetus results in persistently infected (PI) calves being the major risk of BVD spread, but also an efficient target for controlling the infection. There are two acknowledged strategies to identify PI animals for removal: tissue tag testing (direct; also known as the Swiss model) and serological screening (indirect by interpreting the serological status of the herd; the Scandinavian model). Both strategies are effective in reducing PI prevalence and herd incidence. During the first four years of the Irish national BVD eradication programme (2013–16), it has been mandatory for all newborn calves to be tested using tissue tag testing. During this period, PI incidence has substantially declined. In recent times, there has been interest among stakeholders in a change to an indirect testing strategy, with potential benefit to the overall programme, particularly with respect to cost to farmers. Advice was sought on the usefulness of implementing the necessary changes. Here we review available data from the national eradication programme and strategy performance predictions from an expert system model to quantify expected benefits of the strategy change from strategic, budgetary and implementation points of view. Key findings from our work include (i) drawbacks associated with changes to programme implementation, in particular the loss of epidemiological information to allow real-time monitoring of eradication progress or to reliably predict time to eradication, (ii) the fact that only 25% of the herds in the Irish cattle sector (14% beef, 78% dairy herds) would benefit financially from a change to serosurveillance, with half of these participants benefiting by less than EUR 75 per annum at herd level or an average of EUR 1.22 per cow, and (iii) opportunities to enhance the effectiveness of the current programme, particularly in terms of time to eradication, through enforced compliance with PI removal as currently outlined in programme recommendations. The assembled information provides scientific arguments, contributing to an informed debate of the pros and cons of a change in eradication strategy in Ireland.

# Patterns of calving and young stock movement in Ireland and their implications for BVD serosurveillance

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The Republic of Ireland has a national eradication programme for bovine viral diarrhoea virus (BVDv) based on tissue-tag testing of calves to identify persistently infected animals (PIs). It has been proposed that serological testing of a sample of home-bred young stock would be a more cost effective surveillance mechanism than continued tissue-tag testing in herds which have previously been found to be BVD-free. These animals would have to be at least 6 months of age to avoid interference from maternal antibodies in test results. To examine the potential practicality of this system, we identified birth profiles and movements of calves born in Ireland during 2014 and 2015. We found that birth profiles for both beef and dairy animals were more evenly distributed throughout the year than often assumed, which should be borne in mind when evaluating the suitability of a single round of serological testing in the autumn for every herd. A large amount of movement was identified, with approximately 43% of calves experiencing a move before they reached 10 months of age, including moves to another Irish herd, to a knackery, to export, or to slaughter. Approximately 19% of calves had moved to other breeding herds in Ireland within this period. There were distinct patterns according to movement type, month of birth and herd type. The majority of herds moved either all or none of their calves in the first 10 months of life. These results indicate that young stock serological testing is unlikely to be an appropriate surveillance mechanism for all BVDv-free herds, as (i) many herds would not be able to supply a large enough sample of suitably aged home-bred young stock at a single point in time and (ii) PIs which would have been picked up by tissue-tag testing soon after birth would have moved from their home herd, to infect other herds, before serological testing could be conducted.

NON-REGULATORY CATTLE HEALTH ISSUES
Biosecure diseases

## Decision support beyond total saving - benefits for individual cattle herds from changes in monitoring strategy for bovine viral diarrhoea (BVD) in Ireland

Tratalos, J. et al

UCD CVERA

Surveillance and management of livestock diseases is often evaluated with reference to expected sector-wide costs. In contrast, we calculate losses or savings for individual herd owners of a change in monitoring strategy for BVD in Ireland. In this study, we examine the costs faced by each breeding Irish cattle herd if BVD testing was conducted using serology on a sample of young stock, in contrast to the current method of tissue-tag testing of all newborn calves under differing scenarios for the number of management groups. These results will be used to shape future policy in BVD surveillance prior to eradication.

## *Infectious bovine rhinotracheitis (IBR)*

## Expert system modelling of cattle disease management in modern livestock populations and in the context of an Irish IBR eradication programme

Thulke, H.-H. et al.

Helmholtz Centre for Environmental Research - UFZ, Department of Ecological Modelling, Leipzig, Germany

There is a need for expert system modelling to support national policy makers in the design and adaptation of a sustainable national programme to eliminate infectious bovine rhinotracheitis (IBR) from the Irish cattle industry. In this project, mechanistic or process-based dynamic models representing social interaction networks will be developed to better understand how farm-level management characteristics and interaction via trade and market networks affect the local and regional resilience of potential animal health programmes. The model will allow alternative assumptions and strategy options to be tested for their impact on overall programme success.

## *Johne's disease*

## Associations between paratuberculosis ELISA results and test-day records of cows enrolled in the Irish Johne's Disease Control Program

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The effect of the *Mycobacterium avium* ssp. *paratuberculosis* (MAP) ELISA status on test-day milk performance of cows from Irish herds enrolled in the pilot national voluntary Johne's disease control program during 2013 to 2015 was estimated. A data set comprising 92,854 cows and 592,623 complete test-day records distributed across 1,700 herds was used in this study. The resulting ELISA outcome (negative, inconclusive, and positive) of each cow within each year of the program was used to allocate the cow into different scenarios representing the MAP status. At $MAPscenario_1$, all cows testing ELISA nonnegative (i.e., inconclusive and positive) were assigned a MAP-positive status; at $MAPscenario_2$, only cows testing ELISA-positive were assigned a MAP-positive status; at $MAPscenario_3$ only cows testing ELISA nonnegative (inconclusive or positive) and gathered exclusively from herds where at least 2 further ELISA nonnegative (inconclusive or positive) cows were found were assigned a MAP-positive status; at $MAPscenario_4$ only cows testing ELISA-positive that were gathered exclusively from herds where at least 2 further ELISA-positive cows were found were assigned a MAP-positive status. Milk outputs based on test-day records were standardized for fat and protein contents (SMY) and the effect of MAP ELISA status on the SMY was estimated by a linear mixed effects model structure. The SMY mean difference recorded at test day between cows with a MAP-positive status and those with a MAP-negative status within $MAPscenario_1$ was estimated at –0.182 kg/test day; the mean difference was –0.297 kg/test day for $MAPscenario_2$; for $MAPscenario_3$ mean difference between MAP-positive status and MAP test-negative cows was –0.209 kg/test day, and for $MAPscenario_4$, the difference was –0.326 kg/test day.

## NexusMAP

Ezanno, P. et al.

BioEpAR (Biology, epidemiology, and risk analysis in animal health), Nantes, France

For many years, there has been considerable discussion towards the development of a national Johne's disease (JD) control programme for Ireland, in large part to provide assurance to international markets of the safety of Irish livestock products. A national JD model would greatly assist with this, specifically to allow in-depth evaluation of a range of control options. Substantial progress has been made by French colleagues towards a regional JD model, and this project will seek to adapt both a within- and between-herd French model to Irish conditions.

# The effect of paratuberculosis on milk yield – a systematic review and meta-analysis

McAloon, C.G.[1], Whyte. P.[1], More, S.J.[2], Green, M.J.[3], O'Grady, L.[1], Garcia, A.[4], Doherty, M.L.[1]

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Bovine paratuberculosis is a disease characterized by chronic granulomatous enteritis causing protein-losing enteropathy. Adverse effects on animal productivity are key drivers in the attempt to control paratuberculosis at the farm level. Economic models require an accurate estimation of the production effects associated with paratuberculosis. The aim of this study was to conduct a systematic review and meta-analysis to investigate the effect of paratuberculosis on milk production. A total of 20 effect estimates from 15 studies were included in the final meta-analysis. Substantial between-study heterogeneity was observed. Subgroup analysis by case definition and study design was carried out to investigate heterogeneity. The majority of between-study variation was attributed to studies that defined cases on serology. Calculation of a pooled effect estimate was only appropriate for studies that defined cases by organism detection. A reduction in milk yield, corrected for lactation number and herd of origin of 1.87 kg/d, equivalent to 5.9% of yield, was associated with fecal culture or PCR positivity in individual cows.

# Bayesian estimation of prevalence of paratuberculosis in dairy herds enrolled in a voluntary Johne's Disease Control Programme in Ireland

McAloon, C.G.[1], Doherty, M.L.[1], Whyte. P.[1], O'Grady, L.[1], More, S.J.[2], Messam, L.L.McV.[1], Good, M.[3], Mullowney, P.[3], Strain, S.[4], Green, M.J.[5]

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Bovine paratuberculosis is a disease characterised by chronic granulomatous enteritis which manifests clinically as a protein-losing enteropathy causing diarrhoea, hypoproteinaemia, emaciation and, eventually death. Some evidence exists to suggest a possible zoonotic link and a national voluntary Johne's Disease Control Programme was initiated by Animal Health Ireland in 2013. The objective of this study was to estimate herd-level true prevalence (HTP) and animal-level true prevalence (ATP) of paratuberculosis in Irish herds enrolled in the national voluntary JD control programme during 2013–14. Two datasets were used in this study. The first dataset had been collected in Ireland during 2005 (5,822 animals from 119 herds), and was used to construct model priors. Model priors were updated with a primary (2013–14) dataset which included test records from 99,101 animals in 1039 dairy herds and was generated as part of the national voluntary JD control programme. The posterior estimate of HTP from the final Bayesian model was 0.23–0.34 with a 95% probability. Across all herds, the median ATP was found to be 0.032 (0.009, 0.145). This study represents the first use of Bayesian methodology to estimate the prevalence of paratuberculosis in Irish dairy herds. The HTP estimate was higher than previous Irish estimates but still lower than estimates from other major dairy producing countries.

# Relative importance of herd-level risk factors for probability of infection with paratuberculosis in Irish dairy herds

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Control of paratuberculosis is challenging due to the relatively poor performance of diagnostic tests, a prolonged incubation period, and protracted environmental survival. Prioritization of herd-level interventions is not possible because putative risk factors are often not supported by risk factor studies. The objective for this study was to investigate the relative importance of risk factors for an increased probability of herd paratuberculosis infection. Risk assessment data, comprehensive animal purchase history, and diagnostic test data were available for 936 Irish dairy herds. Both logistic regression and a Bayesian β regression on the outcome of a latent class analysis were conducted. Population attributable fractions and proportional reduction in variance explained were calculated for each variable in the logistic and Bayesian models, respectively. Routine use of the calving area for sick or lame cows was found to be a significant explanatory covariate in both models. Purchasing behavior for the previous 10 yr was not found to be significant. For the logistic model, length of time calves spend in the calving pen (25%) and routine use of the calving pen for sick or lame animals (14%) had the highest attributable fractions. For the Bayesian model, the overall $R^2$ was 16%. Dry cow cleanliness (7%) and routine use of the calving area for sick or lame cows (6%) and had the highest proportional reduction in variance explained. These findings provide support for several management practices commonly recommended as part of paratuberculosis control programs; however, a large proportion of the observed variation in probability of infection remained unexplained, suggesting other important risks factors may exist.

NON-REGULATORY CATTLE HEALTH ISSUES
Biosecure diseases

## Johne's disease in the eyes of Irish cattle farmers: a qualitative narrative research approach to understanding implications for disease management

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Bovine Johne's Disease (JD) is a disease characterised by chronic granulomatous enteritis which manifests clinically as a protein-losing enteropathy causing diarrhoea, hypoproteinaemia, emaciation and, eventually death. Some research exists to suggest that the aetiologic pathogen *Mycobacterium avium* subspecies *paratuberculosis* may pose a zoonotic risk. Nationally coordinated control programmes have been introduced in many of the major milk producing countries across the world. However, JD is challenging to control in infected herds owing to limitations of diagnostic tests and the long incubation period of the disease. Internationally, research increasingly recognises that improved understanding of farmers' subjective views and behaviours may inform and enhance disease management strategies and support the identification and implementation of best practice at farm level. The aim of this study was to use qualitative research methods to explore the values and knowledges of farmers relative to the control of JD at farm level. The Biographical Narrative Interpretive Method (BNIM) was used to generate data from both infected and presumed uninfected farms in Ireland. Qualitative analysis revealed that cultural and social capital informed farmers' decisions on whether to introduce control and preventive measures. Cultural capital refers to the pride and esteem farmers associate with particular objects and actions whereas social capital is the value that farmers associate with social relationships with others. On-farm controls were often evaluated by farmers as impractical and were frequently at odds with farmers' knowledge of calf management. Knowledge from farmers of infected herds did not disseminate among peer farmers. Owners of herds believed to be uninfected expressed a view that controls and preventive measures were not worthy of adoption until there was clear evidence of JD in the herd. These findings highlight important barriers and potential aids to prevention and control in both infected and uninfected herds.

## Epidemiology and control of paratuberculosis in dairy herds – a review

McAloon, C. et al.

UCD School of Veterinary Medicine

Bovine paratuberculosis is considered endemic among farmed cattle internationally. There are potential public health implications, given a potential link with Crohn's disease in humans. Milk is considered a potential transmission route to humans and it is recognised that pasteurisation does not necessarily eliminate the bacterium. Therefore, control must also include reduction of the levels of *Mycobacterium avium* spp. *paratuberculosis* supplied from dairy farms. This article reviews the current literature on epidemiology and control of paratuberculosis in dairy herds.

# Non-biosecure diseases, conditions and issues

## *Milk quality*

### The impact of removal of the seasonality formula on the eligibility of Irish herds to supply raw milk for processing of dairy products

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The dairy industry in Ireland is expanding rapidly, with a focus on the production of high quality milk. Somatic cell counts (SCC) are an important indicator both of udder health and milk quality. Milk sold by Irish farmers for manufacture must comply with EU regulations. Irish SCC data is also subject to a monthly seasonal adjustment, for four months from November to February, on account of the seasonality of milk production in Ireland. In a recent study, however, there was no evidence of a dilution effect on SCC with increasing milk yield in Irish dairy cattle. The aim of this paper is to estimate the impact of removal of the seasonality formula on the eligibility of Irish herds to supply raw milk for processing of dairy products. Bulk tank SCC data from 2013 were collected from 14 cooperatives in Ireland. The geometric mean of SCC test results was calculated for each calendar month. We then calculated the number of herds and volume of milk supplied falling in three SCC categories (<200,000, 200,000–400,000, >400,000 cells/mL) in Ireland during 2013 based on their geometric mean SCC every month. Each herd was assigned an 'eligibility to supply' status (always compliant, under warning (first warning, second warning, third warning) and liable for suspension) each month based on their 3-month rolling geometric mean, using methods as outlined in EU and Irish legislation. Two methods were used to calculate the 3-month rolling geometric mean. We then determined the number of herds and volume of milk supplied by 'eligibility to supply' status in Ireland during 2013. All calculations were conducted with and without the seasonality adjustment. The analyses were performed on 2,124,864 records, including 1,571,363 SCC test results from 16,740 herds. With the seasonality adjustment in place, 860 (5.1%) or 854 (5.1%) of herds should have been liable for suspension during 2013 if calculation method 1 or 2, respectively, had been used. If the seasonality adjustment were removed, it is estimated that the number of herds liable for suspension would increase from 860 to 974 (13.2% increase) using calculation method 1, or from 854 to 964 (12.9% increase) using calculation method 2. The modelled impact of such removal would be relatively minor, based on available data, regardless of the method used to calculate the 3-month rolling geometric mean. The focus of the current study was quite narrow, effectively from July to December 2013. Therefore, the results are an underestimate of the total number of herds liable for suspension during 2013. They may also underestimate the true percentage change in herds liable for suspension, with the removal of the seasonality formula. A national herd identifier was lacking from a sizeable percentage of the 2013 bulk tank SCC data, but will be needed if these data are to be meaningfully used for this or other purposes.

# The use of national-level data to describe trends in intramammary antimicrobial usage on Irish dairy farms during 2003 to 2015

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In this study, we used national-level data to describe trends in on-farm intramammary antimicrobial usage in Ireland from 2003 to 2015. We calculated actual sales of intramammary tubes and the quantity of active substance sold, by year, product type [lactation or dry cow therapy (DCT)], antimicrobial group, World Health Organization antimicrobial classification, and from 2009 to 2015, prescribing route. We also estimated on-farm usage of lactation and dry cow intramammary antimicrobials using defined daily dose (DDDvet) and defined course dose (DCDvet) calculations, and dry cow coverage. Sales of tubes of antimicrobial for DCT have increased, and the estimated national dry cow coverage in 2015 was 1,022 DCDvet per 1,000 cows per year. An increase has also occurred in sales of teat sealant (2015 sales: 66.7 tubes with teat sealant for every 100 tubes with antimicrobial for DCT). In contrast, the number of tubes of antimicrobial sold for lactation use has decreased to 1,398 DDDvet and 466 DCDvet per 1,000 animals per year. Sales in intramammary tubes with at least one critically important antimicrobial (CIA) have either risen since 2007 (DCT) or fallen (lactation therapy). Increases were observed in both the number of dry cow and lactation tubes containing CIA considered of highest priority for human health. Differences between prescribing routes with respect to CIA usage were observed. This study provides detailed insight into on-farm usage of intramammary antimicrobials in Ireland. It demonstrates positive national progress but also highlights areas for review. In particular, blanket dry cow treatment in Ireland should be reconsidered. It is not possible to investigate farm-level variation in antimicrobial usage from national sales data. In several countries, measurement and benchmarking have been critical to progress in reducing antimicrobial usage in farm animal production. Central collation of data on farm-level antimicrobial use is also needed in Ireland to allow objective measurement and benchmarking of on-farm usage. More generally, standardized indicators to quantify antimicrobial usage in farm animals are urgently needed to allow country-level comparisons.

## Is there a pathological basis for the observed seasonal rise in somatic cell count in seasonal calving herds?

**Boland, F. et al.**

Population Health Sciences, Royal College of Surgeons in Ireland

As outlined in EU legislation, a seasonality formula is applied to milk supplied in Ireland, allowing somatic cell count (SCC) data to be adjusted in milk supplied during November-February each year. This adjustment assumes a physiological basis for SCC seasonality. However, in earlier work we found no evidence of a dilution effect of SCC with increasing milk yield in Irish dairy cattle. Here, we consider the alternative, whether the seasonal rise in somatic cell count has a pathological basis, and will therefore be more pronounced in animals with a higher average SCC in the current or in the previous lactation. We also consider whether an increase in SCC at the beginning and end of lactation vary for animals with different average SCC in the current and in the previous lactation?

## Mapping milk production in Ireland

**McGrath, G. et al.**

UCD CVERA

This project seeks to present a national map of milk production based on data supplied by the Irish Cattle Breeding Federation for milk recording herds.



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# Other animal health and welfare issues

# Animal welfare

## Conducting sensitive social science research about on-farm animal welfare incidents: challenges and approaches

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The social sciences can help provide a deeper understanding of human-farm animal relations. However, social science research exploring problematic human-farm animal interactions can be of a sensitive nature. Studies that carry risks for participants and the researcher are known methodologically as sensitive research. However, there is little discussion in the animal welfare sciences on how best to conduct research of this nature on animal owners, despite recommendations being made for more interdisciplinary collaboration between the animal welfare sciences and social sciences. Drawing on social science research conducted in 2012 on the human element of on-farm animal welfare incidents in the Republic of Ireland, this short communication presents a case study of the sensitivities and challenges involved in carrying out social science research related to farm animal welfare. This communication details the steps involved in recruiting participants, the methodological challenges encountered, and the approaches used to overcome these challenges. Our experience suggests that when conducting socially sensitive research, careful consideration needs to be applied to the recruitment process, and the study design must aim to minimise the potential risks for all involved. Professionals in the field, such as veterinarians, can play an important role in outlining some of the implications involved, and in overcoming research challenges. Understanding the challenges to this form of research will help to maximise research potential.

## Study on the association between tail lesion score, cold carcass weight and viscera condemnations in slaughter pigs

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The aim of this study was to assess the relationship between tail lesions, cold carcass weight, and viscera condemnations in an Irish abattoir. The following data were collected at the evisceration point from every third pig slaughtered over 7 days: farm identification, sex, tail lesion score, viscera inspection outcome, and cold carcass weight. Tail lesions were scored according to a 5-point scale. Disease lesions responsible for lung (pleurisy, pneumonia, and abscess), heart (pericarditis), and liver (ascariasis) condemnation were recorded based on the decision of the veterinary inspector (VI). Data on 3,143 pigs from 61 batches were available. The relationship between disease lesions, tail lesion score, and cold carcass weight was studied at individual carcass level, while the relationship between disease lesions and tail lesion score was studied at both carcass and batch level. Tail lesions (score ≥1) were found in 72% of the study population, with 2.3% affected by severe tail lesions (scores ≥3). Pleurisy (13.7%) followed by pneumonia (10.4%) showed the highest prevalence, whereas the prevalence of ascariasis showed the greatest variation between batches (0–75%). Tail lesion score, pleurisy, pleuropneumonia, and pericarditis were associated with reductions in carcass cold weight ($P \leq 0.05$) ranging

from 3 to 6.6 kg. Tail lesion score was associated with condemnations for pleurisy, pneumonia, and pleuropneumonia ($P \leq 0.05$) at a batch level. VI shift was associated with condemnations for pneumonia, pleuropneumonia, and pericarditis ($P \leq 0.05$) at a carcass level and with pneumonia at a batch level. Sex was not associated with viscera condemnations but males were more likely to be affected by tail lesions. The relationship between overall tail lesion score and the lung diseases at batch level supports the relationship between poor health and poor welfare of pigs on farms. The inclusion of tail lesion scores at post-mortem meat inspection should be considered as a health and welfare diagnostic tool.

# Transmissible spongiform encephalopathy (TSE)

## Using an epidemiological framework and bovine spongiform encephalopathy investigation questionnaire to investigate suspect bovine spongiform encephalopathy cases: an example from a bovine spongiform encephalopathy case in Ireland in 2015

O'Connor, J.T.[1], Byrne, J.P.[1], More, S.J.[2], Blake, M.[1], McGrath, G.[2], Tratalos, J.A.[2], McElroy, M.C.[1], Kiernan, P.[1], Canty, M.J.[1], O'Brien-Lynch, C.[1], Griffin, J.M.[1]

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In several EU member states, bovine spongiform encephalopathy (BSE) cases have been identified in cattle born after the reinforced ban (BARB cases), for reasons that are not entirely clear. Epidemiological investigation of these cases has proved challenging. The European Food Safety Authority recently recommended the collection of a predefined set of epidemiological data from BSE suspects and confirmed BSE cases to aid future investigations. In this study, we present an epidemiological framework and BSE investigation questionnaire to aid the investigation of suspect BSE cases, and illustrate its application during the investigation of a BSE case in Ireland in 2015. It is recommended that the framework and questionnaire are used concurrently: the framework provides structure and focus, whereas the questionnaire (with 135 questions) aids data collection. The framework focuses on confirmation and discrimination, estimating the date and location of exposure, and determining the method/source of exposure. The BSE case in Ireland in 2015 was a BARB case born in 2010. It was identified with classical BSE at an authorised knackery as part of Ireland's targeted active surveillance programme for BSE. No definitive source of infection with the BSE agent could be attributed in this case.

# Foot and mouth disease

## Enhancement of the current national foot and mouth disease (FMD) spread model

**McGrath, G. et al.**

UCD CVERA

The current FMD model consists of 3 components. An inter-herd disease spread model which controls for virus emission. A wind dispersion model (HYSPLIT) and a risk of infection of model. We intend to revise the first and third component of the overall model to perform better in an Irish context.

## Development of high resolution local scale foot and mouth disease dispersion model

**McGrath, G. et al.**

UCD CVERA

We will investigate the possibility of creating a new dispersion model that will run in conjunction with the existing dispersion model (HYSPLIT). This dispersion model will have a high resolution and will be based on nearest weather station data. This model will offer better precision for potential local spread.

# Schmallenberg virus

## Hypothetical route of the introduction of Schmallenberg virus into Ireland using two complementary analyses

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Ireland lost its official freedom from Schmallenberg virus (SBV) in October 2012. The route of introduction is uncertain, with long-distance displacement of infected *Culicoides*, biting midges, by suitable wind flows considered to be the most likely source. The authors investigated the potential introduction of SBV into Ireland through a *Culicoides* incursion event in the summer of 2012. They conducted SBV serology on archived bovine sera to identify the prospective dispersal window, then used atmospheric dispersion modelling during periods around this window to identify environmental conditions the authors considered suitable for atmospheric dispersal of *Culicoides* from potential infected source locations across Southern England. The authors believe that there was one plausible window over the summer of 2012, on August 10–11, based on suitable meteorological conditions. They conclude that a potential long-range transportation event of *Culicoides* appears to have occurred successfully only once during the 2012 vector competent season. If these incursion events remain at a low frequency, meteorological modelling has the potential to contribute cost-effectively to the alert and response systems for vector borne diseases in the future.
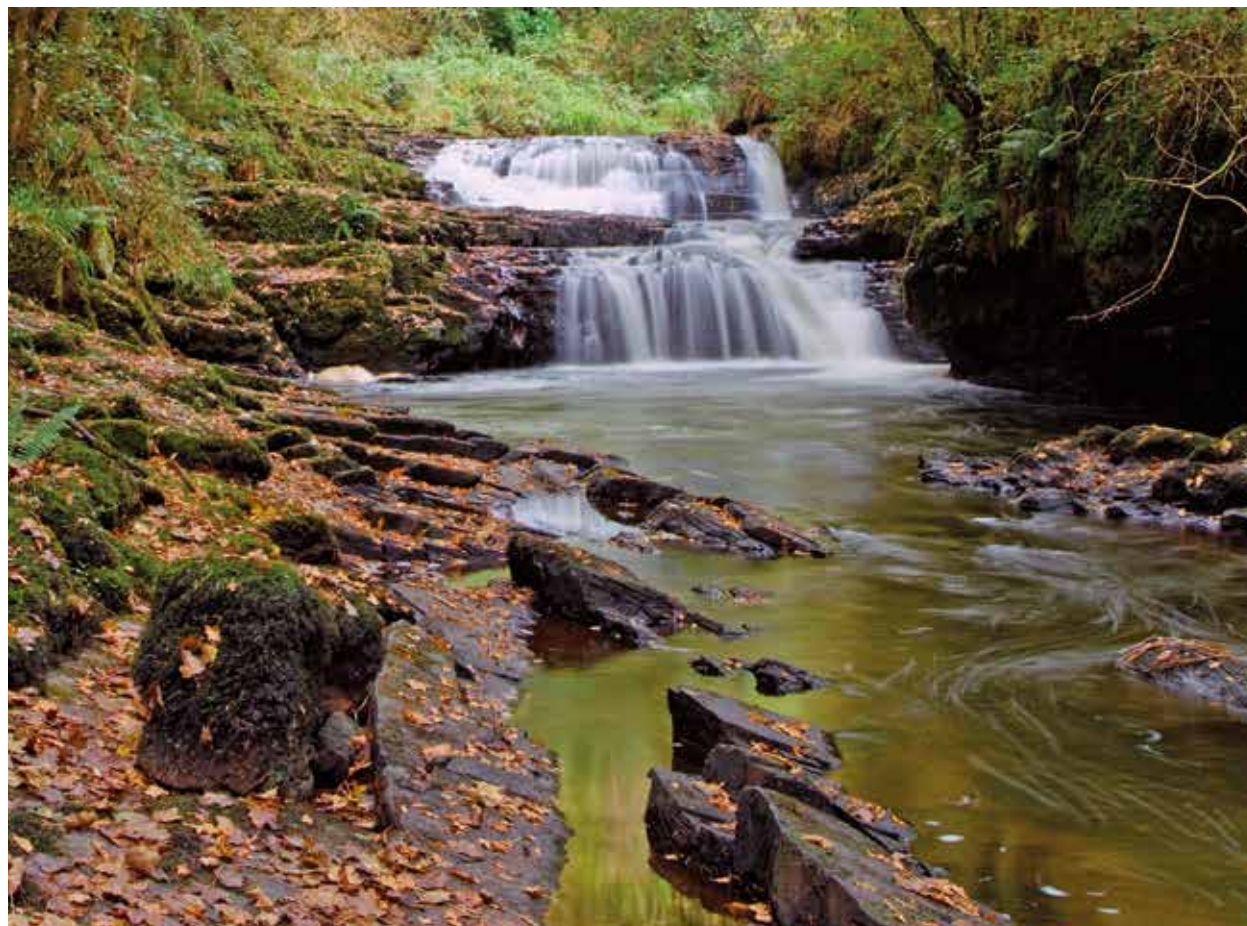
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# Other farmed ruminant health issues

## Influenza D Virus in cattle, Ireland

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We detected influenza D virus in 18 nasal swab samples from cattle in Ireland that were clinically diagnosed with respiratory disease. Specimens were obtained from archived samples received for routine diagnosis during 2014–2016. Sequencing showed that viruses from Ireland clustered with virus sequences obtained in Europe within the D/swine/OK/1334/2011 clade.

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## Risk factors associated with exposure to bovine respiratory disease pathogens during the peri-weaning period in dairy bull calves

Murray, G.M.[1], More, S.J.[2], Clegg, T.[2], Earley, B.[3], O'Neill, R.G.[1], Johnston, D.[3], Gilmore, J.[4], Nosov, M.[4], McElroy, M.C.[1], Inzana, T.J.[5], Cassidy, J.P.[6]

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Bovine respiratory disease (BRD) remains among the leading causes of death of cattle internationally. The objective of this study was to identify risk factors associated with exposure to BRD pathogens during the peri-weaning period (day (d)-14 to d 14 relative to weaning at 0) in dairy bull calves using serological responses to these pathogens as surrogate markers of exposure. Clinically normal Holstein-Friesian and Jersey breed bull calves ($n$ = 72) were group housed in 4

pens using a factorial design with calves of different breeds and planes of nutrition in each pen. Intrinsic, management and clinical data were collected during the pre-weaning (d – 56 to d – 14) period. Calves were gradually weaned over 14 days (d – 14 to d 0). Serological analysis for antibodies against key BRD pathogens (BRSV, BPI3V, BHV-1, BHV-4, BCoV, BVDV and *H. somni*) was undertaken at d – 14 and d 14. Linear regression models (for BVDV, BPI3V, BHV-1, BHV-4, BCoV and *H. somni*) and a single mixed effect random variable model (for BRSV) were used to identify risk factors for changes in antibody levels to these pathogens. BRSV was the only pathogen which demonstrated clustering by pen. Jersey calves experienced significantly lower changes in BVDV S/P than Holstein-Friesian calves. Animals with a high maximum respiratory score (≥8) recorded significant increases in *H. somni* S/P during the peri-weaning period when compared to those with respiratory scores of ≤3. Haptoglobin levels of between 1.32 and 1.60 mg/ml at d – 14 were significantly associated with decreases in BHV-1 S/N during the peri-weaning period. Higher BVDV S/P ratios at d – 14 were significantly correlated with increased changes in serological responses to BHV-4 over the peri-weaning period. Haptoglobin may have potential as a predictor of exposure to BHV-1. BRSV would appear to play a more significant role at the 'group' rather than 'individual animal' level. The significant associations between the pre-weaning levels of antibodies to certain BRD pathogens and changes in the levels of antibodies to the various pathogens during the peri-weaning period may reflect a cohort of possibly genetically linked 'better responders' among the study population.

# A retrospective epidemiological analysis of risk factors for a primary necropsy diagnosis of bovine respiratory disease

Murray, G.M.[1], Cassidy, J.P.[2], Clegg, T.A.[3], Tratalos, J.A.[3], McClure, J.[4], O'Neill, R.G.[1], Sammin, D.J.[1], Casey, M.J.[1], McElroy, M.[1], Earley, B.[5], Bourke, N.[2], More, S.J.[2]

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Bovine respiratory disease (BRD) is a multifactorial disease and the primary cause of both bovine morbidity and mortality in Ireland. The risk factors associated with a primary necropsy diagnosis of BRD among cattle in the traditional (non-feedlot) husbandry systems prevalent in Ireland have not been investigated previously. The aim of this case-control study was to investigate those risk factors among cattle of all ages over an 8 year period. A total of 3,090 BRD cases and 5,236 controls were matched by submitting veterinary practitioner. Univariable and multivariable analyses were performed to examine the association of selected animal-level, herd-level and environmental risk factors with case or control status using a conditional logistical regression model. Male cattle aged more than 31 days were significantly more likely to record a primary necropsy diagnosis of BRD than female cattle. Older cattle of both sexes were at increased odds of a BRD necropsy diagnosis than younger calves with the exception of female cattle aged greater than 165 days. The risk of a primary necropsy diagnosis of BRD increased with increasing herd size and decreased with increasing time in days since the last animal movement into the submitting herd. There were significantly reduced odds of a primary necropsy diagnosis of BRD in the summer (June to August) when compared with the autumn (September to November). These findings identify significant risk factors for a necropsy diagnosis of BRD under non-feedlot-type husbandry conditions.

## Evolving views on bovine respiratory disease: An appraisal of selected key pathogens – Part 1

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Bovine respiratory disease (BRD) is one of the most commonly diagnosed causes of morbidity and mortality in cattle and interactions of factors associated with the animal, the pathogen and the environment are central to its pathogenesis. Emerging knowledge of a role for pathogens traditionally assumed to be minor players in the pathogenesis of BRD reflects an increasingly complex situation that will necessitate regular reappraisal of BRD pathogenesis and control. This review appraises the role of selected key pathogens implicated in BRD pathogenesis to assess how our understanding of their role has evolved in recent years.

## Evolving views on bovine respiratory disease: An appraisal of selected control measures – Part 2

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Bovine respiratory disease (BRD) control poses significant challenges to the cattle industry worldwide. The sometimes complex interactions of factors associated with the animal, the pathogen and the environment complicate the implementation of effective control measures. Blanket vaccination or mass medication provides inconsistent control and the effective tackling of BRD will require innovative, evidence-based and targeted interventions which, if employed sensibly, offer useful alternatives for addressing this disease. This review appraises the role of the specific interventions employed in BRD control to assess how our understanding of their role and efficacy has evolved in recent years.

**OTHER ANIMAL HEALTH AND WELFARE ISSUES**
Other farmed ruminant health issues

# Pathogens, patterns of pneumonia, and epidemiologic risk factors associated with respiratory disease in recently weaned cattle in Ireland

Murray, G.M.[1], More, S.J.[2], Sammin, D.[1], Casey, M.J.[1], McElroy, M.C.[1], O'Neill, R.G.[1], Byrne, W.J.[1], Earley, B.[3], Clegg, T.A.[2], Ball, H.[4], Bell, C.J.[4], Cassidy, J.P.[5]

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We examined the pathogens, morphologic patterns, and risk factors associated with bovine respiratory disease (BRD) in 136 recently weaned cattle ("weanlings"), 6–12 mo of age, that were submitted for postmortem examination to regional veterinary laboratories in Ireland. A standardized sampling protocol included routine microbiologic investigations as well as polymerase chain reaction and immunohistochemistry. Lungs with histologic lesions were categorized into 1 of 5 morphologic patterns of pneumonia. Fibrinosuppurative bronchopneumonia (49%) and interstitial pneumonia (48%) were the morphologic patterns recorded most frequently. The various morphologic patterns of pulmonary lesions suggest the involvement of variable combinations of initiating and compounding infectious agents that hindered any simple classification of the etiopathogenesis of the pneumonias. Dual infections were detected in 58% of lungs, with *Mannheimia haemolytica* and *Histophilus somni* most frequently recorded in concert. *M. haemolytica* (43%) was the most frequently detected respiratory pathogen; *H. somni* was also shown to be frequently implicated in pneumonia in this age group of cattle. *Bovine parainfluenza virus 3* (BPIV-3) and *Bovine respiratory syncytial virus* (16% each) were the viral agents detected most frequently. Potential respiratory pathogens (particularly *Pasteurella multocida*, BPIV-3, and *H. somni*) were frequently detected (64%) in lungs that had neither gross nor histologic pulmonary lesions, raising questions regarding their role in the pathogenesis of BRD. The breadth of respiratory pathogens detected in bovine lungs by various detection methods highlights the diagnostic value of parallel analyses in respiratory disease postmortem investigation.

# The bovine paranasal sinuses: bacterial flora, epithelial expression of nitric oxide and potential role in the in-herd persistence of respiratory disease pathogens

Murray, G.M.[1], O'Neill, R.G.[1], Lee, A.[2], McElroy, M.[1], More, S.J.[3], Monagle, A.[2], Earley, B.[4], Cassidy, J.P.[2]

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The bovine paranasal sinuses are a group of complex cavernous air-filled spaces, lined by respiratory epithelium, the exact function of which is unclear. While lesions affecting these sinuses are occasionally reported in cattle, their microbial flora has not been defined. Furthermore, given that the various bacterial and viral pathogens causing bovine respiratory disease (BRD) persist within herds, we speculated that the paranasal sinuses may serve as a refuge for such infectious agents. The paranasal sinuses of clinically normal cattle (n = 99) and of cattle submitted for post-mortem examination (PME: n = 34) were examined by microbial culture, PCR and serology to include bacterial and viral pathogens typically associated with BRD: *Mycoplasma bovis, Histophilus somni, Mannheimia haemolytica* and *Pasteurella multocida*, bovine respiratory syncytial virus (BRSV) and bovine parainfluenza-3 virus (BPIV-3). Overall, the paranasal sinuses were either predominantly sterile or did not contain detectable microbes (83.5%: 94.9% of clinically normal and 50.0% of cattle submitted for PME). Bacteria, including BRD causing pathogens, were identified in relatively small numbers of cattle (<10%). While

serology indicated widespread exposure of both clinically normal and cattle submitted for PME to BPIV-3 and BRSV (seroprevalences of 91.6% and 84.7%, respectively), PCR identified BPIV-3 in only one animal. To further explore these findings we investigated the potential role of the antimicrobial molecule nitric oxide (NO) within paranasal sinus epithelium using immunohistochemistry. Expression of the enzyme responsible for NO synthesis, inducible nitric oxide synthase (iNOS), was detected to varying degrees in 76.5% of a sub-sample of animals suggesting production of this compound plays a similar protective role in the bovine sinus as it does in humans.

## Antibodies to *Coxiella burnetii* in Irish bulk tank milk samples

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*Coxiella burnetii* is the aetiological agent of Q fever, a zoonotic disease with a worldwide distribution whose main reservoirs are goats, sheep and cattle. In ruminants, infection can cause abortion and reproductive problems, with large numbers of infectious organisms shed at parturition, in milk and in urine and faeces. Infection of humans occurs mainly via the respiratory route, with airborne spread possible beyond the boundaries of the infected farm. In Ireland, there were six recorded human cases of Q fever in humans in 2016, but it is reasonable to assume that the true human incidence is considerably higher. Infection with *C burnetii* is common in Irish cattle herds, with previous research estimating a herd prevalence of 37.9 per cent and an animal level prevalence of 1.8 percent. The Irish dairy sector is currently expanding following the abolition of EU milk quotas, and the trend indicates a likely expansion of the national dairy herd for some years to come. This short communication set out to obtain a more accurate estimate of the prevalence of antibodies to *C burnetii* in Irish dairy herds at this transitional point.

OTHER ANIMAL HEALTH AND WELFARE ISSUES
Other farmed ruminant health issues

# Veterinary ethics

## Ethical challenges facing veterinary professionals in Ireland: results from Policy Delphi with vignette methodology

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Ethics is key to the integrity of the veterinary profession. Despite its importance, there is a lack of applied research on the range of ethical challenges faced by veterinarians. A three round Policy Delphi with vignette methodology was used to record the diversity of views on ethical challenges faced by veterinary professionals in Ireland. Forty experts, comprising veterinary practitioners, inspectors and nurses, accepted to participate. In round 1, twenty vignettes describing a variety of ethically challenging veterinary scenarios were ranked in terms of ethical acceptability, reputational risk and perceived standards of practice. Round 2 aimed at characterising challenges where future policy development or professional guidance was deemed to be needed. In round 3, possible solutions to key challenges were explored. Results suggest that current rules and regulations are insufficient to ensure best veterinary practices and that a collective approach is needed to harness workable solutions for the identified ethical challenges. Challenges pertaining mostly to the food chain seem to require enforcement measures whereas softer measures that promote professional discretion were preferred to address challenges dealing with veterinary clinical services. These findings can support veterinary representative bodies, advisory committees and regulatory authorities in their decision making, policy and regulation.

## Challenges facing the veterinary profession in Ireland: 1. Clinical veterinary services

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The provision of veterinary clinical services is known to elicit a range of challenges which require an ethical appraisal. In a recent Policy Delphi study, referrals/second opinions and 24 h emergency care were identified as matters of key concern by veterinary professionals in Ireland. In this case study (the first in a series of three resulting from a research workshop exploring challenges facing the veterinary profession in Ireland; the other two case studies investigate the on-farm use of veterinary antimicrobials and emergency/casualty slaughter certification) we aim to provide a value-based reflection on the constraints and possible opportunities for two prominent veterinary clinical services in Ireland: referrals/second opinions and 24 h emergency care. Using a qualitative focus group approach, this study gathered evidence from relevant stakeholders, namely referral and referring veterinarians, clients, animal charities, and the regulatory body. Six overarching, interrelated constraints emerged from the thematic analysis: the need to improve current guidance, managing clients' expectations, concerns with veterinarian well-being, financial issues, timeliness of referral, and conflicts between veterinary practices. Possible solutions to improve veterinary referral and out-of-hours clinical services included clarifying the terms used in current norms and regulations (namely 'referral', 'second opinion', '24 h emergency care' and '24 h cover'), improved communication (making the client aware of the different levels of veterinary care that are being offered, and transparent and full disclosure of clinical records), and the promotion of Continuing Veterinary Education

in communication, business management and ethical decision-making. These findings may help inform the Veterinary Council of Ireland about future recommendations and regulatory measures.

# Challenges facing the veterinary profession in Ireland: 2. On-farm use of veterinary antimicrobials

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Antimicrobial resistance has emerged in recent years as a significant public health threat, which requires both an ethical and a scientific approach. In a recent Policy Delphi study, on-farm use of antimicrobials was a key concern identified by veterinary professionals in Ireland. In this case study (the second in a series of three resulting from a research workshop exploring the challenges facing the veterinary profession in Ireland; the other two case studies investigate clinical veterinary services and emergency/casualty slaughter certification) we aim to provide a value-based reflection on the constraints and possible opportunities for responsible use of veterinary antimicrobials in Ireland. Using a qualitative focus group approach, this study gathered evidence from relevant stakeholders, namely veterinarians working in public and private organisations, a representative from the veterinary regulatory body, a dairy farmer and a general medical practitioner. Three overarching constraints to prudent on-farm use of veterinary antimicrobials emerged from the thematic analysis: 'Defective regulations', 'Lack of knowledge and values' regarding farmers and vets and 'Farm-centred concerns', including economic and husbandry concerns. Conversely, three main themes which reflect possible opportunities to the barriers were identified: 'Improved regulations', 'Education' and 'Herd health management'. Five main recommendations arose from this study based on the perspectives of the study participants including: a) the potential for regulatory change to facilitate an increase in the number of yearly visits of veterinarians to farms and to implement electronic prescribing and shorter validity of prescriptions; b) a 'One Health' education plan; c) improved professional guidance on responsible use of veterinary antimicrobials; d) improved on-farm herd health management practices; and e) the promotion of a 'One Farm-One Vet' policy. These findings may assist Veterinary Council of Ireland and other competent authorities when revising recommendations concerning the prudent use of veterinary antimicrobials in farmed animals.

OTHER ANIMAL HEALTH AND WELFARE ISSUES
Veterinary ethics

# Challenges facing the veterinary profession in Ireland: 3. Emergency and casualty slaughter certification

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Veterinarians are faced with significant conflicts of interest when issuing certificates for the transport and slaughter of acutely injured and casualty livestock. In a recent Policy Delphi study, emergency and casualty slaughter certification was a key concern identified by veterinary professionals in Ireland. In this case study (the third in a series of three resulting from a research workshop exploring challenges facing the veterinary profession in Ireland; the other two case studies investigate clinical veterinary services and the on-farm use of veterinary antimicrobials), we aim to provide a value-based reflection on the constraints and opportunities for best practice in emergency and casualty slaughter certification in Ireland. Using a qualitative focus group approach, this study gathered evidence from relevant stakeholders, namely a representative from the regulatory body, local authority veterinarians with research experience in emergency slaughter, an animal welfare research scientist, official veterinarians from the competent authority, a private veterinary practitioner, and a member of a farming organisation. Results revealed a conflict between the responsibility of private veterinary practitioners (PVPs) to safeguard the welfare of acutely injured bovines on-farm and the client's commercial concerns. As a consequence, some PVPs may feel under pressure to certify, for example, an acutely injured animal for casualty slaughter instead of recommending either on-farm emergency slaughter or disposal by the knackery service. Among Official Veterinarians, there are concerns about the pressure within processing plants to accept acutely injured livestock as casualty animals. Confusion pertaining to legislation and definition of fitness to travel also contribute to these dilemmas. Conflicts of interest arise due to the gap between governance and provision to facilitate on-farm emergency slaughter of livestock. Increased availability and acceptance of on-farm emergency slaughter by Food Business Operators (FBOs) would mitigate the need to certify acutely injured animals fit for transport and slaughter and thereby safeguard animal welfare. In the absence of nationwide availability and acceptance of on-farm emergency slaughter by FBOs, consideration should be given to methods to encourage all those involved in the food chain to prioritise animal welfare when in conflict with the commercial value of the animal. Training and guidelines for PVPs on the regulatory landscape and ethical decision-making should become available. The reintroduction of the fallen animal scheme should be considered to support farm animal welfare.

# Marine animal health

## Mortality in *Crassostrea gigas* oysters in Ireland during 2012

Morrissey, T.[1], Houtsma, E.[2], Cheslett, D.[1], Yatabe, T.[3], Geoghegan, F.[1], More, S.J.[2]

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OsHV-1 µVar-related mortality is well described in several countries, including France. In Ireland, however, clear patterns in relation to the incidence and impact of this virus have been difficult to identify, at least in part because seawater temperatures rarely exceed 16°C, a recognised threshold for OsHV-1 µVar-related mortality, for prolonged periods. In this study, based on data collected during 2012, we aimed to evaluate mortality levels and the prevalence of OsHV-1 µVar in *C. gigas* oysters in Ireland. This was done by comparing oyster stocks from different sources grown under similar management and environmental conditions within the same bay and oyster stocks grown under different management and environmental conditions between bays. Wilcoxon Rank Sum Test was used to evaluate differences in the mean mortality of oysters and a negative binomial regression model was used to evaluate differences in OsHV-1 µVar prevalence between and within bays. A visual assessment of temperature changes was conducted during the period when mortality events were first recorded. Factors that showed evidence of some association with *C. gigas* mortality during 2012 include temperature increase and hatchery source. However, no consistent association between *C. gigas* oyster mortality and OsHV-1 µVar prevalence during 2012 was observed, either within or between bays. The results presented in this study, though conflicting in part, reflect the presentation of OsHV-1 µVar related oyster mortalities observed in Ireland over the past number of years.

## Can biosecurity and network properties predict pathogen species richness in the salmonid industry?

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Salmonid farming in Ireland is mostly organic, which implies limited disease treatment options. This highlights the importance of biosecurity for preventing the introduction and spread of infectious agents. Similarly, the effect of local network properties on infection spread processes has rarely been evaluated. In this paper, we characterized the biosecurity of salmonid farms in Ireland using a survey, and then developed a score for benchmarking the disease risk of salmonid farms. The usefulness and validity of this score, together with farm indegree (dichotomized as ≤ 1 or > 1), were assessed through generalized Poisson regression models, in which the modeled outcome was pathogen richness, defined here as the number of different diseases affecting a farm during a year. Seawater salmon (SW salmon) farms had the highest biosecurity scores with a median (interquartile range) of 82.3 (5.4), followed by freshwater salmon (FW salmon) with 75.2 (8.2), and freshwater trout (FW trout) farms with 74.8 (4.5). For FW salmon and trout farms, the top ranked model (in terms of leave-one-out information criteria, looic) was the null model (looic = 46.1). For SW salmon farms,

## Co-modelling the dynamics of the network of live fish movements in Ireland and disease transmission

Yatabe, T. et al.

Centre for Animal Disease Modelling and Surveillance (CADMS), Dept. Medicine & Epidemiology, School Veterinary Medicine,
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In earlier work, we characterized the live salmonid movement network in Ireland, and identified three locations as hotspots of highly central sites with a higher potential for both introduction and spread of infection. Subsequently, we characterized the biosecurity of Irish salmonid farms, and noted the importance of biosecurity and farm centrality (specifically indegree) in predicting pathogen species richness. Here, we co-model the dynamics of the network of live fish movements in Ireland and disease transmission, using as a case study a disease of concern to the Irish salmonid farms.

OTHER ANIMAL HEALTH AND WELFARE ISSUES
Marine animal health

# Food safety & quality

## An outbreak of *Salmonella* Enteritidis in broiler chickens in Ireland, 2015

Aznar, I. et al.

UCD CVERA & DAFM

Salmonellosis is a worldwide food-borne disease of major public health concern and the second most common zoonosis in the European Union. *Salmonella enterica* serovar Enteritidis (*Salmonella* Enteritidis) was the most frequently reported serovar in cases of human Salmonellosis in the EU in 2014 and 2015 accounting for 44.4% and 45.7% respectively of all salmonella cases. Human infections with this serovar are frequently associated with poultry via the intake of infected eggs and egg products. This project will describe two outbreaks of *Salmonella* Enteritidis in broiler flocks in Ireland during 2015 which involved two different strains: PT8 and PT21. Details of the larger of these two outbreaks (PT21) including the most likely source of infection and timeline events will be described. Infected farms will be described in time and space and lessons learned during the investigation will be discussed.

## Private animal health and welfare standards in quality assurance programmes: a review and proposed framework for critical evaluation

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In recent years, 'private standards' in animal health and welfare have become increasingly common, and are often incorporated into quality assurance (QA) programmes. Here, we present an overview of the use of private animal health and welfare standards in QA programmes, and propose a generic framework to facilitate critical programme review. Private standards are being developed in direct response to consumer demand for QA, and offer an opportunity for product differentiation and a means to drive consumer choice. Nonetheless, a range of concerns have been raised, relating to the credibility of these standards, their potential as a discriminatory barrier to trade, the multiplicity of private standards that have been developed, the lack of consumer input and compliance costs. There is a need for greater scrutiny of private standards and of associated QA programmes. We propose a framework to clarify the primary programme goal(s) and measureable outputs relevant to animal health and welfare, the primary programme beneficiaries and to determine whether the programme is effective, efficient and transparent. This paper provides a theoretical overview, noting that this framework could be used as a tool directly for programme evaluation, or as a tool to assist with programme development and review.

**OTHER ANIMAL HEALTH AND WELFARE ISSUES**
Food safety & quality

# Efficacy of washing and disinfection in cattle markets in Ireland

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Few studies have reported on the effectiveness of the washing and disinfection methods used in cattle markets in Ireland. Purchasing cattle into recipient herds poses a high biosecurity risk due to the possibility of introducing disease. In Ireland, livestock markets are an important intermediary in the movement of cattle to new herds. Thus disease control strategies need to consider the disease risk associated with moving livestock through markets. Some cattle are also moved directly from markets for slaughter at abattoirs. Washing and disinfection at markets is utilised to reduce faecal contamination in markets, thereby reducing the risk of disease spread among animals and carcass contamination at slaughter. The primary objective of this study was to assess the efficacy of standard washing and disinfection techniques at markets in Ireland in reducing bacterial contamination on internal structures. Total viable counts (TVC) of colony forming units (CFU) were used as indicators of bacterial contamination, which could include pathogens of public and animal health concern. Four hundred and seventy nine samples were taken mainly from pen floors and the TVC enumerated for each sample. Washing and disinfection was effective at significantly reducing TVCs on floors and metal bars of market holding pens, but residual contamination remained. Washing market pens only (no disinfection), followed by a rest period between batches of cattle (6.5 days) was as effective at reducing TVCs as washing followed by disinfection and a shorter rest period (5.5 days). Markets are a potential reservoir for microbial contamination with a resultant increased risk of disease spread by cattle moving through markets into new herds, and carcass contamination for cattle moving directly to slaughter. Therefore, market managers need clear advice and guidance on the development of hygiene programmes that are suitable for use in livestock markets.

**OTHER ANIMAL HEALTH AND WELFARE ISSUES**
Food safety & quality

# Tick-borne diseases

## Ectoparasites and associated diseases in sheep and cattle in Ireland; ticks as vectors of disease

**Good, B. et al.**

Teagasc

This project aims to determine the current significance of ticks, mites and lice and the effectiveness of control measures used on Irish farms. The importance of ticks as vectors of disease will be comprehensively evaluated alongside the efficacy of current control measures.

## Current and future risk associated with ticks and tick-borne diseases: research and surveillance

**Zintl, A. et al.**

UCD School of Veterinary Medicine

This project aims to carry out a survey of tick species and their distribution on the island of Ireland. The data will be used to develop predictive maps for endemic tick species and TBDs and assess how their distributions are likely to be affected by various climate change scenarios. The likely survival and establishment of exotic tick and pathogens species will also be considered. Finally, novel analytical tools will be developed which combine stable isotope ratio and multiplex molecular analyses and facilitate simultaneous identification of TBPs and important reservoir hosts.



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# Miscellaneous

## Bluetongue monitoring

Tratalos, J. et al

UCD CVERA

The aim of this project is to provide evidence of continued freedom from disease for Bluetongue Virus in Ireland, using blood samples from the Irish cull cow programme. A stratified spatial sampling strategy is used, with the number of samples from each of 51 grid squares measuring 45 km$^2$ each in Ireland taken in proportion to the number of bovines resident in the grid square. To fit with the biting season of the *Culicoides* midge (the principal vector of the disease) only animals which have remained in the same locality from 1st April till the day before slaughter are used. Computer code allows the selection of the geographically stratified samples from the cull cow sample blocks whilst minimising the number of blocks which need to be thawed.

## Understanding the context for pet cat and dog feeding and exercising behaviour among pet owners in Ireland: a qualitative study

Downes, M.J.[1, 2, 3], Devitt, C.[4], Downes, M.T.[1], More, S.J.[3]

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Pet cat and dog obesity contributes to increased risk of several diseases, including cancer and diabetes mellitus as well as a worsening of orthopaedic problems, and a reduction in survival rate. This study aims to develop a better understanding of cat and dog owners' self-reported beliefs and factors that influence owner behaviour around feeding and exercising their pet cat or dog, as there is a lack of in-depth understanding in this area. Seven focus group discussions, with 43 pet owners in total, were conducted. Pet owners often reported a perceived a low level of control over feeding; often undermined by other people feeding of their pet, their pets begging for food, and their pets attitude towards food. Treats were used in the absence of owner control over pet begging and emotional attachment, and to influence pet behaviour. The majority of participants had positive attitudes to pet exercise, which could be related to pet specific requirements, especially differences in cats and dogs. There were some negative experiences of stress associated with dog walking and fears over aggressive confrontations with other dogs. Feeding one's pet is influenced by beliefs about pet specific needs, pet food and pet health, pet owners' perceived control over feeding, and the implications for the pet owner. Pet exercise is influenced by beliefs about pet specific exercise needs, and the implications of exercising one's pet for the pet owner. Understanding owner behaviours on feeding and exercise allows for a more targeted approach to preventing and treating pet obesity.

# Key factors affecting reproductive success of thoroughbred mares and stallions on a commercial stud farm

Lane, E.A.[1,2], Bijnen, M.L.J.[3], Osborne, M.[4], More, S.J.[2], Henderson, I.S.F.[5], Duffy, P.[6], Crowe, M.A.[3]

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To evaluate factors contributing to fertility of thoroughbred mares, data from 3,743 oestrous periods of 2,385 mares were collected on a large thoroughbred farm in Ireland. Fourteen stallions (mean age 8.3 years; range 4–15 years) had bred 2,385 mares (mean age 9.4 years; range 3–24 years). Maiden mares accounted for 12%, mares with a foal at foot for 64%, and barren, slipped or rested mares for 24% of the total. The mean pregnancy rate per cycle was 67.8% (68.6% in year 1 and 66.9% in year 2). Backward stepwise multivariable logistic regression analysis was utilized to develop two models to evaluate mare factors, including mare age, reproductive status, month of foaling, dystocia, month of cover, foal heat, cycle number, treatments, walk-in status and stallion factors including stallion identity, stallion age, shuttle status, time elapsed between covers and high stallion usage on the per cycle pregnancy rate and pregnancy loss. Old age ($p < 0.001$) and cover within 20 days post-partum ($p < 0.003$) were associated with lowered pregnancy rates. High mare age ($p < 0.05$) and barren, slipped or rested reproductive status ($p = 0.05$) increased the likelihood of pregnancy loss. Uterine inflammation or infection, if appropriately treated, did not affect fertility. Only high usage of stallions (used more than 21 times in previous week) was associated with lowered ($p = 0.009$) pregnancy rates. However, shuttle stallions were more likely to have increased ($p = 0.035$) pregnancy survival, perhaps reflecting a bias in stallion selection. In conclusion, mare age exerted the greatest influence on fertility; nonetheless, thoroughbreds can be effectively managed to achieve high reproductive performance in a commercial setting.

# European cattle movement networks

Colizza, V. et al.

Inserm, Paris, France

This collaborative study, conducted between researchers from 15 European countries, is comparing European cattle trade markets to examine similarities and specific characteristics of each and to assess the implications for infectious disease spread, public health and animal welfare. This study will allow us to see how Ireland differs from its neighbours, and in turn what this says about our vulnerability to disease and how best to conduct monitoring programmes. This involves running generic computer code on Irish movement data in CVERA and sharing the resulting metrics with the larger project, but no sharing of the raw data.

## Geographical distribution of *Angiostrongylus vasorum* in foxes *(Vulpes vulpes)* in the Republic of Ireland

McCarthy, G.[1], Ferrand, M.[2], DeWaal, T.[1], Zintl, A.[1], McGrath, G.[3], Byrne, W.[4], O'Neill, E.J.[1]

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[3] UCD CVERA, [4] DAFM Veterinary Laboratory Service

The reported incidence of the metastrongylid nematode *Angiostrongylus vasorum*, that infects dogs and other canids, is increasing worldwide outside recognized endemic foci. This apparent expansion of the parasite's range is causing concern to veterinary clinicians as the disease caused in dogs can be life threatening and its treatment is not straightforward. The red fox is thought to be a reservoir host for dogs. To investigate the spatial distribution of infection in foxes in Ireland, the hearts and lungs of 542 foxes from all over Ireland were examined. The incidence of infection was found to be 39·9% [95% confidence interval (CI) 35·7–44·1] with positive samples occurring in each of the country's 26 counties. This report confirms that the parasite is endemic in Ireland and the overall prevalence is the second highest in Europe. This is the first survey of *A. vasorum* infection in Irish foxes and highlights the potential exposure of the Irish dog population to high risk of cross-infection. Additionally, *Crenosoma vulpis* was found in seven of the foxes, a parasite not previously reported in the Irish fox.

## Live meteorological forecasting model for *Culicoides* incursion risk

McGrath, G. et al.

UCD CVERA

Met Éireann and CVERA collaborated to produce a live operational model showing the predicted dispersion of *Culicoides* from a number of locations in Northern France and south and southwest England. Department of Agriculture Food and the Marine staff receive daily outputs from the model which includes 1 day of observation data and 3 days of forecast data. The model is currently very basic and requires more physics and biological parameters to be added to make it more realistic. On completion, the model will act as a guide for when and where to target bulk milk testing for incursion of Bluetongue virus. The model will also be used with historical meteorological data to assess the frequency that high risk events occur in Ireland.

## Develop a working model for predicting and identifying the locations of contaminated farms following a nuclear accident event

McGrath, G. et al.

UCD CVERA

CVERA, in conjunction with the Office of Radiological Protection (ORP), Met Éireann and the Department of Agriculture, Food and the Marine, will create a model for defining the extent and severity of contamination of farm land both during and after a nuclear accident scenario in the United Kingdom or continental Europe. A risk value will be assigned to all active herds/flocks which will be appraised by DAFM in their contingency management.


## Co-infection study within EU-funded PARAGONE (H2020) project

Mulcahy, G. et al.

UCD School of Veterinary Medicine

This project aims to investigate whether there is a correlation (at individual- and herd-level) between fasciolosis and other important endemic diseases, with particular emphasis on paramphistomosis and Johne's disease.


## An investigative framework to facilitate epidemiological thinking during herd problem-solving

More, S.J.[1], Doherty, M.L.[2], O'Grady, L.[2]
[1] UCD CVERA, [2] UCD School of Veterinary Medicine

Veterinary clinicians and students commonly use diagnostic approaches appropriate for individual cases when conducting herd problem-solving. However, these approaches can be problematic, in part because they make limited use of epidemiological principles and methods, which has clear application during the investigation of herd problems. In this paper, we provide an overview of diagnostic approaches that are used when investigating individual animal cases, and the challenges faced when these approaches are directly translated from the individual to the herd. Further, we propose an investigative framework to facilitate epidemiological thinking during herd problem-solving. A number of different approaches are used when making a diagnosis on an individual animal, including pattern recognition, hypothetico-deductive reasoning, and the key abnormality method. Methods commonly applied to individuals are often adapted for herd problem-solving: 'comparison with best practice' being a herd-level adaptation of pattern recognition, and 'differential diagnoses' a herd-level adaptation of hypothetico-deductive reasoning. These approaches can be effective, however, challenges can arise. Herds are complex; a collection of individual cows, but also additional layers relating to environment, management, feeding etc. It is unrealistic to expect seamless translation of diagnostic approaches from the individual to the herd. Comparison with best practice is time-consuming and prioritisation of actions can be problematic, whereas differential diagnoses can lead to 'pathogen hunting', particularly in complex cases. Epidemiology is the science of understanding disease in populations. The focus is on the population, underpinned by principles and utilising methods that seek to allow us to generate solid conclusions from apparently uncontrolled situations. In this paper, we argue for the inclusion of epidemiological principles and methods as an additional tool for herd problem-solving, and outline an investigative framework, with examples, to effectively incorporate these principles and methods with other diagnostic approaches during herd problem-solving. Relevant measures of performance are identified, and measures of

## Research impact in animal health and welfare: examples from Ireland

More, S.J. et al.

UCD CVERA

There is a growing body of literature focusing on the concept of effectiveness at the science-policy interface, specifically those factors that maximise the usefulness of scientific knowledge in influencing policy decision-making. This issue has been considered in a number of contexts, including public health and global environmental issues such as climate change. As yet, there is limited information on factors influencing effectiveness at the science-policy interface in animal health and welfare. This paper will address this issue, drawing on lessons in animal health and welfare from Ireland.

## Association between rumen fluke and liver fluke prevalence in ruminants in Ireland

Naranjo Lucena, A. et al.

UCD School of Veterinary Medicine

This study will look at the spatial variation in presence/absence and co-infection of liver and rumen fluke in Ireland.

# Risk assessment of pesticides and other stressors in bees: Principles, data gaps and perspectives from the European Food Safety Authority

Rortais, A.[1], Arnold, G.[2], Dorne, J.-L.[1], More, S.J.[3], Sperandio, G.[4], Streissl, F.[1], Szentes, C.[1], Verdonck, F.[1]

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Current approaches to risk assessment in bees do not take into account co-exposures from multiple stressors. The European Food Safety Authority (EFSA) is deploying resources and efforts to move towards a holistic risk assessment approach of multiple stressors in bees. This paper describes the general principles of pesticide risk assessment in bees, including recent developments at EFSA dealing with risk assessment of single and multiple pesticide residues and biological hazards. The EFSA Guidance Document on the risk assessment of plant protection products in bees highlights the need for the inclusion of an uncertainty analysis, other routes of exposures and multiple stressors such as chemical mixtures and biological agents. The EFSA risk assessment on the survival, spread and establishment of the small hive beetle, *Aethina tumida*, an invasive alien species, is provided with potential insights for other bee pests such as the Asian hornet, *Vespa velutina*. Furthermore, data gaps are identified at each step of the risk assessment, and recommendations are made for future research that could be supported under the framework of Horizon 2020. Finally, the recent work conducted at EFSA is presented, under the overarching MUST-B project ("EU efforts towards the development of a holistic approach for the risk assessment on MUltiple STressors in Bees") comprising a toolbox for harmonised data collection under field conditions and a mechanistic model to assess effects from pesticides and other stressors such as biological agents and beekeeping management practices, at the colony level and in a spatially complex landscape. Future perspectives at EFSA include the development of a data model to collate high quality data to calibrate and validate the model to be used as a regulatory tool. Finally, the evidence collected within the framework of MUST-B will support EFSA's activities on the development of a holistic approach to the risk assessment of multiple stressors in bees. In conclusion, EFSA calls for collaborative action at the EU level to establish a common and open access database to serve multiple purposes and different stakeholders.

## Multi-criteria decision analysis to model *Ixodes ricinus* habitat suitability

Rousseau, R.[1], McGrath, G.[2], McMahon, B.J.[3], Vanwambeke, S.O.[1]

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[2] UCD CVERA, [3] UCD School of Agriculture and Food Science

Tick-borne diseases present a major threat to both human and livestock health throughout Europe. The risk of infection is directly related to the presence of its vector. Thereby it is important to know their distribution, which is strongly associated with environmental factors: the presence and availability of a suitable habitat, of a suitable climate and of hosts. The present study models the habitat suitability for *Ixodes ricinus* in Ireland, where data on tick distribution are scarce. Tick habitat suitability was estimated at a coarse scale (10 km) with a multi-criteria decision analysis (MCDA) method according to four different scenarios (depending on the variables used and on the weights granted to each of them). The western part of Ireland and the Wicklow mountains in the East were estimated to be the most suitable areas for *I. ricinus* in the island. There was a good level of agreement between results from the MCDA and recorded tick presence. The different scenarios did not affect the spatial outputs substantially. The current study suggests that tick habitat suitability can be mapped accurately at a coarse scale in a data-scarce context using knowledge-based methods. It can serve as a guideline for future countrywide sampling that would help to determine local risk of tick presence and refining knowledge on tick habitat suitability in Ireland.

## VKORC1 sequence variants associated with resistance to anticoagulant rodenticides in Irish populations of *Rattus norvegicus* and *Mus musculus domesticus*

Mooney, J.[1], Lynch, M.R.[2], Prescott, C.V.[3], Clegg, T.[4], Loughlin, M.[5], Hannon, B.[6], Moore, C.[7], Faulkner, R.[7]

[1] DAFM Veterinary Laboratory Service, [2] Campaign for Responsible Rodenticide Use Ireland CLG, [3] School of Animal and Microbial Sciences, The University of Reading, Reading, United Kingdom, [4] UCD CVERA, [5] Emel Consulting, [6] Ecolab Ireland, [7] Rentokil Initial Ltd.

While resistance to anticoagulant rodenticides is known to occur in many European populations of Norway rat and house mouse, to-date no data is available on the occurrence in Ireland of such resistance. No genetic evidence for the occurrence of resistance was found in 65 Norway rat samples analysed, indicative of an absence, or low prevalence, of resistance in rats in at least the Eastern region of the island of Ireland. The presence of two of the most commonly found amino acid substitutions Leu128Ser and Tyr139Cys associated with house mouse resistance to anticoagulant rodenticides was confirmed. The occurrence of two such mutations is indicative of the occurrence of resistance to anticoagulant rodenticides in house mice in the Eastern region of the island of Ireland.
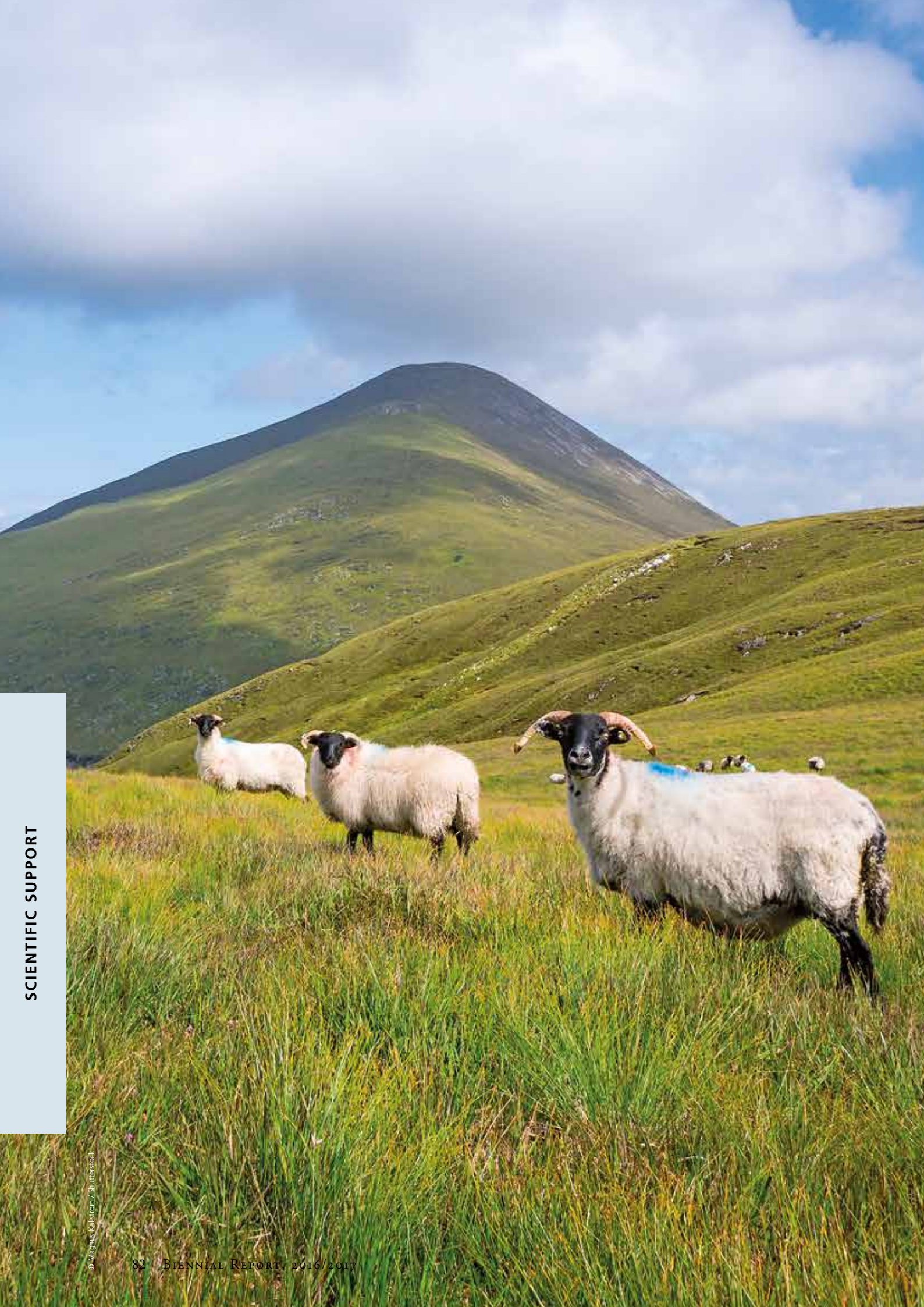
SCIENTIFIC SUPPORT

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# Scientific support

SCIENTIFIC SUPPORT

# Scientific support

CVERA works to generate scientific information in support of national policy decision-making. This work is conducted in two ways, either as:
- defined *scientific projects* (as outlined in earlier sections of this Biennial Report [Bovine tuberculosis; Non-regulatory cattle health issues; Other animal health and welfare issues]), or
- as more-general *scientific support.*

Each accounts for approximately 50% of available resources.

CVERA provides scientific support to both *ad hoc* or ongoing activities, and to a range of national bodies including the Department of Agriculture, Food and the Marine, University College Dublin and Animal Health Ireland. In each case, the work draws on expertise within CVERA in epidemiology, statistics, geographic information systems and database maintenance and interrogation.

The following provide a broad, non-exhaustive overview of scientific support provided by CVERA during 2016 and 2017.

# Epidemiological support

## Simon More & Inma Aznar

### Department of Agriculture, Food and the Marine
- Member, Scientific Advisory Committee on Animal Health and Welfare
- Member, Surveillance Steering Group
- Member, Early Warning System working group
- Member, Abattoir surveillance working group
- Member, bTB/BR North/South Working Group
- Member, Interdepartmental AMR Consultative Committee
- Epidemiological support on a range of issues including:
  - Design of national animal disease surveillance, including a national bluetongue survey to substantiate freedom
  - Measurement of antimicrobial usage in farmed animals
  - Characteristing animal movement: implications for national disease control
  - Rapid risk assessment tools for animal disease outbreaks
  - Development of a risk prioritization method for disease control

### University College Dublin
Teaching
- Undergraduate, including Agricultural Science (ANSC30130), Veterinary Medicine (VETS30170, VETS30290) and Medicine (MDSA10210)
- Postgraduate, including Agricultural Science (AESC40020) and Dairy Herd Health (VETS40180)

Postgraduate supervision/support
- Finalised during 2016/17: Racem Ben Romdhane (PhD École Nationale Vétérinaire, Agroalimentaire et de l'Alimentation, Nantes-Atlantique) [paratuberculosis epidemiology]
- Ongoing: Inma Aznar (PhD Wageningen) [bTB epidemiology], Damien Barrett (PhD) [Schmallenberg epidemiology], Ger Murray (PhD) [bovine respiratory disease], Fiona Reardon (MVSc) [BVD epidemiology], Tadaishi Yatabe (PhD UC Davis) [fish disease epidemiology]

## Other

- Animal Health Ireland
  - ❍ Chair of the Technical Working Groups on Johne's disease and mastitis
  - ❍ Provide scientific support to AHI on issues relating to the BVD, Johne's disease, CellCheck and IBR programmes
- European Food Safety Authority
  - ❍ Chair of the Animal Health and Welfare (AHAW) Panel
  - ❍ Member of the Scientific Committee (SC)
  - ❍ Chair of the MUST-B (multiple stressors on honey bee colony health) working group
  - ❍ Member of the African Swine Fever (ASF) working group
- Food Safety Authority of Ireland
  - ❍ Member of the Scientific Committee
- FP7 RISKSUR project
  - ❍ Membership of the Scientific Advisory Board member
- General scientific community
  - ❍ Scientific Advisory Board, *Preventive Veterinary Medicine*
  - ❍ Deputy Editor, *Irish Veterinary Journal*
  - ❍ Independent referee for a number of international peer reviewed journals
  - ❍ Thesis examination (MVSc, PhD, *Doctor Medicinae Veterinariae*), several universities
  - ❍ Presentation at AFBI 2018 Science Outlook Conference
  - ❍ Member, Organising and Scientific Committees for the Seventh International Conference on *Mycobacterium bovis* (*M. bovis* 2020)

# Statistical support

## Tracy Clegg & Erik Houtsma

## Department of Agriculture, Food and the Marine

- Statistical support on a range of issues including:
  - ❍ National bTB eradication programme
  - ❍ Non-regulatory diseases such as bovine viral diarrhoea
  - ❍ Member, Abattoir surveillance working group
  - ❍ Estimation of the potency of tuberculins against the Irish and International standards
  - ❍ Presentation to DAFM and AHI staff as part of a surveillance workshop
  - ❍ Presentations as part of DAFM's Veterinary Inspector training programme
  - ❍ Statistical advice on a study looking at Johne's disease diagnostic tests
  - ❍ Assistance with the study design for a national liver fluke survey in sheep
  - ❍ Selection of bulk milk samples for a bluetongue and Q fever survey

## University College Dublin

- Statistical support for a range of studies, including:
  - ❍ Teixeira et al, 2016. Study on the association between tail lesion score, cold carcass weight and viscera condemnations in slaughter pigs. *Frontiers in Veterinary Science* 3, 24
  - ❍ Hogan et al, 2016. Optimisation of the zinc sulphate turbidity test for the determination of immune status. *Veterinary Record* 178, 169
  - ❍ Magalhães-Sant'Ana et al, 2016. Ethical challenges facing veterinary professionals in Ireland: results from Policy Delphi with vignette methodology. *Veterinary Record* 179, 437
  - ❍ Bree et al, 2018. Prevalence and risk factors for hyperthyroidism in Irish cats from the greater Dublin area. *Irish Veterinary Journal* 71, 2

## Other
- Statistical support for a range of studies, including:
  - Risk factors for failing the six-month check test for bovine tuberculosis of de-restricted Irish cattle herds, E. Steenbergen [Wageningen University]
  - Independent referee for a number of international peer reviewed journals

# Geographic Information Systems (GIS) support

### Guy McGrath, Daniel M. Collins & Jamie Tratalos

## Department of Agriculture, Food and the Marine

Wildlife Administration Unit

- DAFM's Wildlife Administration Unit (WAU) has had responsibility for the implementation of the Department's wildlife strategy for bovine tuberculosis since its foundation in 2002. The strategy is implemented under licence from, and in co-operation with, National Parks and Wildlife Service (NPWS). CVERA provides support to the WAU as it progresses to the badger vaccination phase of the TB eradication programme, as announced by the Minister for Agriculture, Food and the Marine, Michael Creed, TD in January, 2018. CVERA will continue to provide:
  - Daily approvals and monthly/yearly reports
  - End of year progress maps for each DVO
  - Area treated calculations are submitted on a regular basis in compliance with NPWS
  - Resources for problem areas
  - Resources for the vaccine areas and the non-inferiority trial
  - Identification of setts suitable for bait marking studies
  - Liaising with Coillte for data sharing on setts locations
  - Provision of data to the National Biodiversity Data Centre
  - Review microchip technology
- Provide GIS support for the development of the DAFM "TB mapper"
- Provide quarterly TB mapping reports
- Generate annual "farms in catchment" output for DAFM
- Revise current operational BTV dispersion model to include biological parameters
- GIS for Annual Bluetongue freedom from disease analysis
- GIS support for:
  - Murray, G.M., Cassidy, J.P., Clegg, T.A., Tratalos, J.A., McClure, J., O'Neill, R.G., Sammin, D.J., Casey,M.J.,McElroy, M., Earley, B., Bourke, N., More, S.J., 2016. A retrospective epidemiological analysis of risk factors for a primary necropsy diagnosis of bovine respiratory disease. *Preventive Veterinary Medicine* 132, 49-56
  - Murray, G.M., More, S.J., Sammin, D., Casey, M.J., McElroy, M.C., O'Neill, R.G., Byrne, W.J., Earley, B., Clegg, T.A., Ball, H., Bell, C.J., Cassidy, J.P., 2017. Pathogens, patterns of pneumonia, and epidemiologic risk factors associated with respiratory disease in recently weaned cattle in Ireland. *Journal of Veterinary Diagnostic Investigation* 29, 20-34
- GIS assistance for Central Veterinary Laboratories studies:
  - Produce porcine effluent discharge estimates for waterways to facilitate targeted testing of shellfish for Hep. E
  - General spatial analysis
  - Silvermines investigation
  - Assistance in CBA report for Regional Veterinary Laboratories

## University College Dublin

- GIS support for a range of studies, including:
  - ❍ Grant application to DAFM for National Tick Survey
  - ❍ Rumen/liver fluke studies
  - ❍ Whole Genome Sequencing (WGS) of *Mycobacterium bovis* from cattle and badgers to identify if cross infection is occurring

## Other

- Animal Health Ireland
  - ❍ Provide monthly BVD maps
  - ❍ Additional *ad hoc* support for AHI driven projects and education programmes
- GIS support for:
  - ❍ Graham, D.A., Gallagher, C., Carden, R.F., Lozano, J.-M., Moriarty, J., O'Neill, R., 2017. A survey of free-ranging deer in Ireland for serological evidence of exposure to bovine viral diarrhoea virus, bovine herpes virus-1, bluetongue virus and Schmallenberg virus. *Irish Veterinary Journal* 70, 13
- Member of Irish HYSPLIT dispersion model working group
- Support as European panel expert for the World Meteorological Organisation for Agromet products and services

# Database and computer programming support

## Jamie Tratalos

## Department of Agriculture, Food and the Marine

- AIM Bovine Statistics Report 2015 & 2016
- Annual Bluetongue freedom from disease analysis
- Foot and mouth dispersion modelling
- Provision of bovine movement datasets for use in a variety of projects
- Creation of herd and animal level TB datasets for use by CVERA staff

## Other

- Database support for a range of studies, including:
  - ❍ Development of SQL data bases to ensure that CVERA is in compliance with national Data Protection legislation and that data is readily available in standardised formats for analysis by CVERA staff
  - ❍ Creation of database for Badger Vaccine Field Trial

PUBLICATIONS

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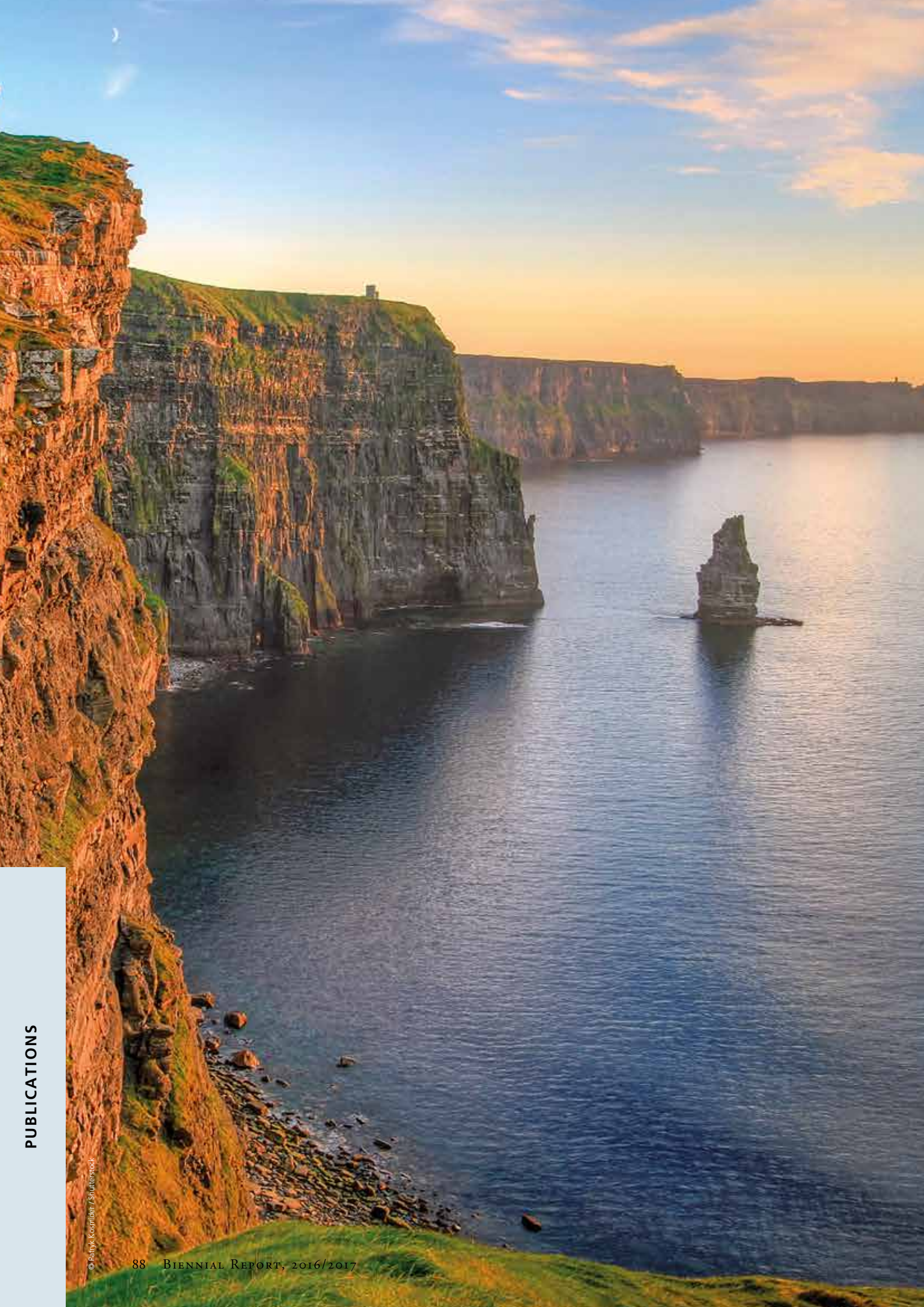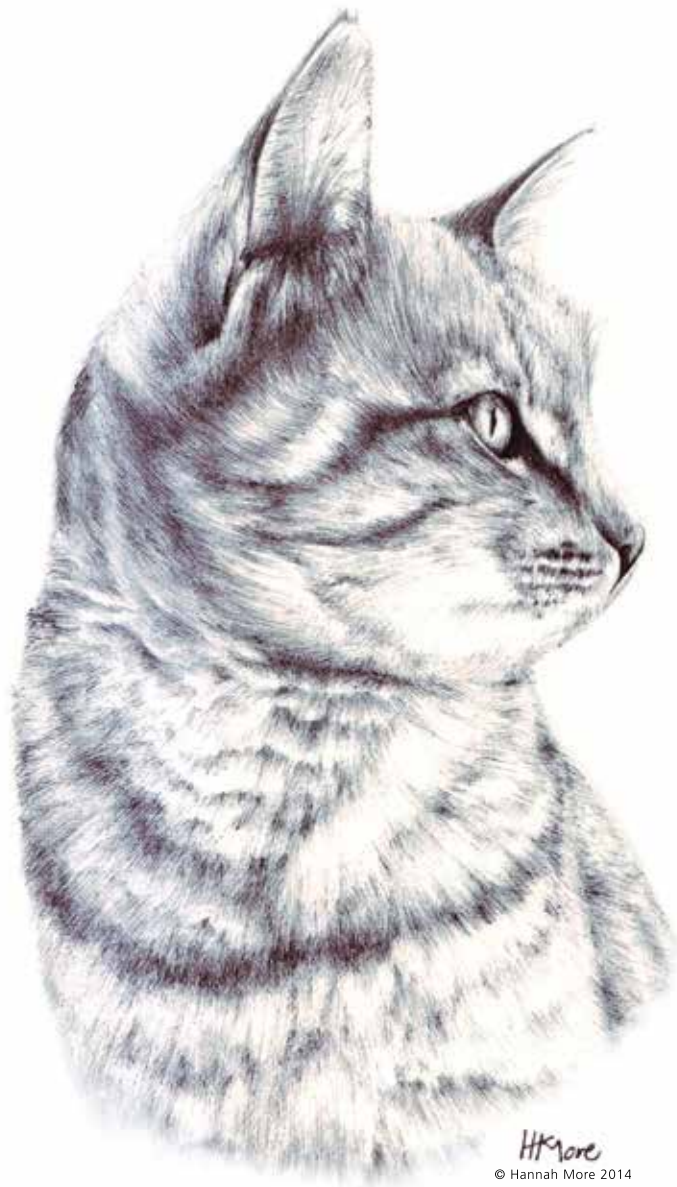
# Publications

# During 2016 - 2017

## *Peer reviewed papers & book chapters*

Abdou, M., Frankena, K., O'Keeffe, J., Byrne, A.W., 2016. Effect of culling and vaccination on bovine tuberculosis infection in a European badger *(Meles meles)* population by spatial simulation modelling.
*Preventive Veterinary Medicine* 125, 19-30.

Botaro, B.G., Ruelle, E., More, S.J., Strain, S., Graham, D.A., O'Flaherty, J., Shalloo, L., 2017. Associations between paratuberculosis ELISA results and test-day records of cows enrolled in the Irish Johne's Disease Control Program.
*Journal of Dairy Science* 100, 7468-7477.

Byrne, A.W., Do Linh San, E., 2016. A cautionary note on the use of Minimum Number Alive-Derived Trappability Metrics in wildlife programmes, as exemplified by the case of the European Badger *(Meles meles)*.
*Wildlife Biology in Practice* 12, 51-57.

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*Frontiers in Cellular and Infection Microbiology* 7, 6.

Clegg, T.A., Good, M., Doyle, M., Duignan, A., More, S.J., Gormley, E., 2017. The performance of the interferon gamma assay when used as a diagnostic or quality assurance test in *Mycobacterium bovis* infected herds. *Preventive Veterinary Medicine* 140, 116-121.

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Clegg, T.A., Graham, D.A., O'Sullivan, P., McGrath, G., More, S.J., 2016. Temporal trends in the retention of BVD+ calves and associated animal and herd-level risk factors during the compulsory eradication programme in Ireland.
*Preventive Veterinary Medicine* 134, 128-138.

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Fenlon, C., O'Grady, L., McCoy, F., Houtsma, E., More, S.J., 2017. The impact of removal of the seasonality formula on the eligibility of Irish herds to supply raw milk for processing of dairy products.
*Irish Veterinary Journal* 70, 9.

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*Preventive Veterinary Medicine* 126, 30-38.

Lane, E.A., Bijnen, M.L.J., Osborne, M., More, S.J., Henderson, I.S.F., Duffy, P., Crowe, M.A., 2016. Key factors affecting reproductive success of thoroughbred mares and stallions on a commercial stud farm. *Reproduction in Domestic Animals* 51, 181-187.

Magalhães-Sant'Ana, M., More, S.J., Morton, D.B., Hanlon, A., 2016. Ethical challenges facing veterinary professionals in Ireland: results from Policy Delphi with vignette methodology. *Veterinary Record* 179, 437.

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McAloon, C., Macken-Walsh, A., Moran, L., Whyte, P., More, S.J., O'Grady, L., Doherty, M.L., 2017. Johne's disease in the eyes of Irish cattle farmers: a qualitative narrative research approach to understanding implications for disease management. *Preventive Veterinary Medicine* 141, 7-13.

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McAloon, C.G., Doherty, M.L., Whyte. P., O'Grady, L., More, S.J., Messam, L.L., Good, M., Mullowney, P., Strain, S., Green, M.J., 2016. Bayesian estimation of prevalence of paratuberculosis in dairy herds enrolled in a voluntary Johne's Disease Control Programme in Ireland. *Preventive Veterinary Medicine* 128, 95-100.

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