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G5- Veterinary Programmes

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REPORT OF THE

**“Bovine Brucellosis”
TASK FORCE SUB-GROUP**

**Meeting held in
Newry
Northern Ireland, UK
06-07 December 2011**

**REPORT OF THE
MEETING OF THE BOVINE BRUCELLOSIS SUB-GROUP OF THE
TASK FORCE FOR MONITORING DISEASE ERADICATION
HELD IN NEWRY, NORTHERN IRELAND, UK, 06-07 DECEMBER 2012**

PARTICIPANTS: see Annex I

AGENDA: see Annex II

LOCATION: Newry, Northern-Ireland, UK

DAY 1

1. Introduction and overview

Owen Denny

The Newry Divisional Veterinary Office, Glenree House, Carnbane Industrial Estate, Newry was chosen as the location of this meeting. Mr. Owen Denny, Senior Principal Veterinary Officer (SPVO) of the Enzootics Section gave an overview of eradication programme on Bovine Brucellosis (BB) and the current situation.

The Veterinary Service forms part of the Department of Agriculture and Rural Development (DARD), Northern Ireland (NI), and is divided in 2 Divisions: the Enzootic Disease & Animal Welfare Division with 3 Sections and the Epizootic Disease & Veterinary Public Health Division with 6 Sections. NI is divided into 10 Divisional Veterinary Office (DVO) areas. The total number of personnel is approximately 600, including 48 Veterinary officers (VOs) and 120 Animal Health & Welfare Inspectors (AHWIs). They have to deal with a total bovine population of 1.6 M and 1.8 M sheep in NI.

The history of BB eradication in NI, the evolution of herd and animal incidence and prevalence during the last 15 years was presented (Fig.1).

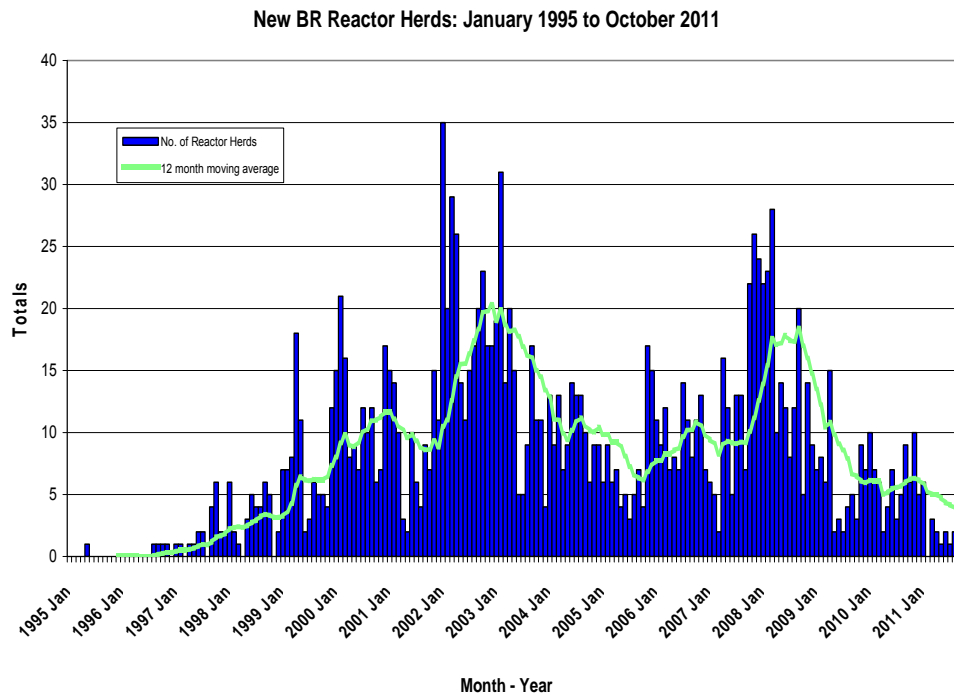


Fig.1: Number of Brucellosis reactor herds

The recommendations and measures implemented since the last Task Force visit in October 2008 in NI were discussed. Furthermore, the measures not or partially implemented were analysed as were the actions and initiatives that contributed to progress made during the last three years.

A total of 9 herds are currently under restriction due to confirmed infection, of which 7 were depopulated totally and one partially. As of 14 July 2011, there have been no new confirmed outbreaks; therefore the Official Services are cautiously optimistic about the potential to achieve eradication of BB in NI.

2. Epidemiology – the current picture in NI

Darrell Abernethy

Mr. Abernethy commenced with a description of brucellosis trends in N. Ireland since 1991 (see Figure below). At that time the country was largely free of the disease, and the herd incidence remained low until 1996. Factors that contributed to the increase in brucellosis from 1996 to 2002 included significant spread from several primary outbreaks, diversion of resources from the eradication programme to deal with the BSE crisis and exotic disease outbreaks (Newcastle disease, 1996; Foot and Mouth Disease, 2001), and latent infection in cattle (resulting in outbreaks some years after exposure and contact testing). Following the FMD epidemic, additional resources and measures were introduced and these resulted in a decline in incidence until the present time. However, a localised cluster in County Armagh in 2005, arising from one badly affected herd, and two small clusters in 2009, led to an increased incidence for several years following the outbreaks.

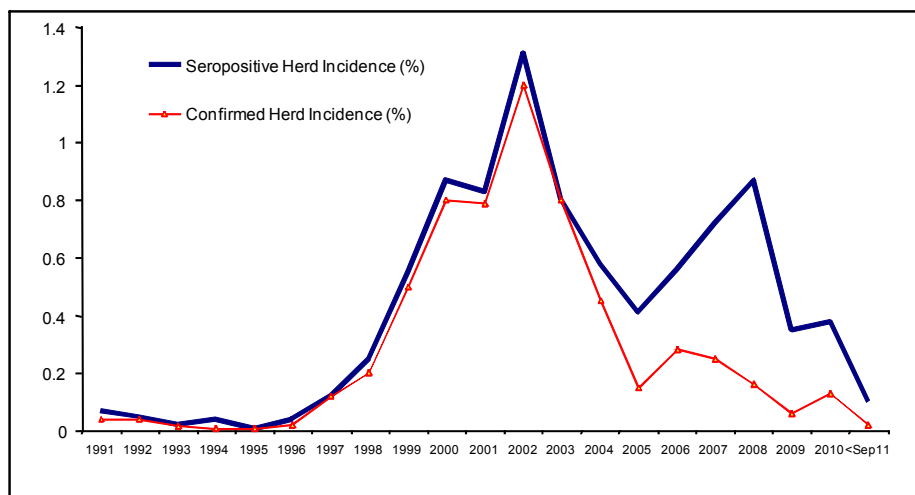


Fig. 2: Annual Herd Incidence in Northern Ireland, 1991 to Sep. 2011

In order to improve the sensitivity of the tests used in the programme, test thresholds were reduced from 2003 and parallel testing introduced from 2005. These measures reduced the specificity of the test regime with the result that the seropositive herd incidence was significantly higher than the culture-positive incidence.

Several studies have been conducted in Northern Ireland to assess risk factors for bovine brucellosis, the most important of which was found to be exposure to infected cattle in the immediate neighbourhood. Approximately 60% of breakdowns occurred in herds that were in close proximity to existing outbreaks, hence the emphasis on intensive and extended testing of neighbouring herds. Other risk factors included herd size, a history of infection or potential exposure, abortions in the herd and purchase of cattle.

It has become clear over the duration of the current epidemic that different types of outbreaks occur. The most common are those that follow natural spread from a primary or secondary outbreak; these follow “normal” epidemiological patterns and are resolved through the current testing regime. However, a small proportion of outbreaks occur through illegal activity, either the deliberate infection of cattle for compensatory gain or accidental transmission of infection through illegal movement of cattle. Examples of both these types of activity have been detected in recent years and it is likely that their relative importance has grown as the epidemic has declined.

3. Northern Ireland Brucellosis Scheme

Sharon Verner

The Veterinary Service (VS) is the Competent Authority under EC Directive 64/432 and legislative authority for the Scheme is based on the Brucellosis Control Order (Northern Ireland) 2004 and the Brucellosis (Examination and Testing) Scheme Order (NI) 2004. The programme uses a test-and-slaughter policy as vaccination against brucellosis is prohibited. It includes the following: individual identification and movement control of all bovines, computerised management of the scheme, involvement of all stakeholders in testing and collaboration with human health authorities and the Health & Safety Executive.

Responsibility for policy rests with the Animal Health and Welfare Policy Division of DARD, while VS delivers the programme. The country is divided into 10 areas and further subdivided into patches with technical officers of VS responsible for all testing, although Private Veterinary Practitioners (PVPs) and private lay testers (working under the supervision of a PVP) may be approved by DARD to carry out blood sampling for pre-movement testing. In 2008 a Brucellosis Programme Management team was established to coordinate all aspects of monitoring and management within the programme. Laboratory testing is carried out at Veterinary Sciences Division (VSD), part of the Agri-Food and Biosciences Institute (AFBI), Northern Ireland.

Herds and cattle may be subject to movement restrictions due to a positive or inconclusive blood test result, suspicion of infection on the holding, clinical symptoms (abortions) or putative contact with infected cattle. Movement controls can be applied to individual animals, herds or to areas ("Temporary Control Zones").

Blood samples are tested using serum agglutination test (SAT) in accordance with EU 64/432 Annex C. If any SAT reading exceeds 30 IU, the sample is retested using an SAT test and complement fixation test (CFT). Any animal giving an SAT test result of > 30 IU of agglutination per ml or any CFT reading of < 20 IU is classified as an inconclusive reactor and is required to be isolated and retested. Risk analysis is carried out: if significant risk factors exist, then an ELISA test is requested on subsequent tests. Testing may be classified as surveillance, used for early detection of the presence of spread of infection, and targeted testing, used where brucellosis is suspected.

Surveillance includes the following:

(1) Herd Testing:

Annual serological testing of all herds is conducted in three divisional areas (DVOS) with a historical higher incidence. They include Armagh, Newry and Enniskillen. In the other seven DVOs, non-dairy herds are tested annually while dairy herds (20 % of cattle population) are blood-tested biennially and

monthly by bulk milk testing. All breeding and potential breeding cattle over 12 months old are tested.

(2) Bulk Milk Testing:

Monthly testing of all dairy herds commenced in 2001. Approx. 38,000 milk samples were tested in 2010; 4.5 % of positive herds have been detected historically through this method.

(3) Abattoir Sampling:

This includes testing of all individually-restricted animals, all cows over 48 months (from 2009) and now, over 72 months.

(4) Laboratory submissions:

All samples (blood, tissue, foetuses) submitted to AFBI are screened for brucellosis. In 2010, 800 cases were tested.

(5) Pre-movement testing:

This has been compulsory since 2004 for all non-castrated cattle over 12 months old. A 30- day interval is used. In 2010, approx. 167,000 tests were conducted and 45,000 samples collected at herd-level tests; 6 reactors were disclosed. Pre-movement testing has had the effect of dampening movement.

Targeted Testing includes:

(a) Post-abortion testing:

All bovine abortions must be reported to DARD. In 2010, 2,600 animals were so tested.

(b) Risk Testing:

This may be undertaken in reactor herds, herds neighbouring outbreaks, traced animals or in Temporary Control Areas.

(c) Brucellin Skin Testing:

This was a recommendation of the 2008 EU Task Force but it has taken several years to obtain the appropriate authorisations. A certificate was issued in March 2011 and trial testing has been undertaken.

Following disclosure of a positive blood test result, the herd is restricted and the reactor isolated prior to slaughter. If *Brucella abortus* is confirmed through culture, the herd is depopulated unless there are compelling economic or risk-based reasons for not doing so. Adjoining farmers are alerted and their herds are restricted until two (inner ring) or one (outer ring) negative tests have been conducted. The herds are tested at least every 3 months during the outbreak. Control measures following disclosure of infection include forward and backward tracing, cleansing and disinfection, slaughter of reactor progeny, prohibition on re-stocking for six months, extended testing after re-stocking, treatment of slurry, sampling of sheep if present and epidemiological investigations.

Valuation of stock to be slaughtered is conducted on-farm; reactors are compensated at 75 % of the estimated value of the animal or 75 % of the average market value, whichever is the lesser. Contact animals contacts are compensated at 100 % value.

Communication with a wide variety of stakeholders is undertaken, consisting of advertising, farmers' meetings liaison groups, as well as meetings with medical colleagues.

Future plans include refinement of parallel testing and surveillance, development of a non-specific (reactors) policy and a communication strategy.

4. Compensation update

Ian McKee

The NI compensation system was updated in 2002 and is based on farm valuation. Reactors are compensated at 75 % of the estimated value of the animal or 75 % of the average market value, whichever is the lesser. Contact animals are compensated at 100 % the market value. There is no ceiling on the amount paid. The salvage value of the carcasses is paid to the Competent Authority.

During the BB Task Force meeting of October 2008 the compensation scheme was questioned and analysed. The Task Force was concerned that the current compensation scheme in NI could encourage fraud and could also encourage farmers not to implement bio-security measures in a correct way. Therefore, the 2008 visit recommended a review of the compensation scheme and a ceiling be set for compensation. The compensation levels used in other MS should also be considered (Report - SANCO/4233/2008).

In 2009, the NI Assembly's Public Accounts Committee's "Report on the Control of Bovine Tuberculosis in Northern Ireland" queried whether:

- 100 % compensation provided a sufficient incentive for herd owners to prevent infection
- it was appropriate that almost 100 % of the costs of animal disease compensation is borne by the taxpayer.

In order to review the compensation scheme in NI, public consultations were held in February - April 2011 and in October - December 2011. The proposals were:

- to continue to value animals on an individual basis
- to limit compensation by applying a cap (ceiling)
- to reduce compensation for contact animals to 75 % (same level applied to reactors).

5. Headquarters management of brucellosis breakdowns

Judith Graham

When a brucellosis outbreak occurs, the first role of the Headquarters (HQ) VO is to receive the notification by the Veterinary Officer (VO), assess the details about the outbreak and advise on immediate actions to be taken. The HQ VO also coordinates all communications about the outbreaks to the appropriate sections involved. To quality assure existing procedures, a dedicated check-list has been developed during the last two years to ensure that both the tasks to be carried out in the field and by HQ are covered. The main information included in the check-list are taken from the Brucellosis database, the APHIS system and from custom made spreadsheets. Early actions for the HQ VO are to coordinate DNA traceability of reactors, check their removal is completed in due time (15 days) and verify that details held in the APHIS system are correct. The HQ VO also checks if appropriate restrictions have been put in place in the herd concerned and in the herds related (adjacent or traced) and that the required tests and notices have been set by the Field VO. Later checks are made to quality assure the full reporting of outbreak details by the VO, notification of depopulation decision, implementing forward and backward traces, recording of the strain isolated and the need for a visit from Veterinary Epidemiology Unit or the need for a DNA comparison. Final tasks are connected to publicity and communications, such as raising awareness of general public and farmers on brucellosis issues using press articles, stakeholder meetings, media (radio/TV), and practical items with brucellosis messages.

6. Veterinary Services Enforcement Branch

Danny Gray

The aim of the Veterinary Services Enforcement Branch (VSEB) is to supervise farmers' respect of existing rules on brucellosis prevention and control. The Branch is structured in two divisions: the Central Enforcement Team (specialised in complex and confrontational cases) and the Divisional

Enforcement Team (for straightforward and less confrontational cases). The VSEB selects the cases to be investigated after an initial evaluation which takes into account different aspects, such as the evidence of the offence, the opportunity of prosecution in the view of the public interest, the resources available to obtain evidence, the opportunity for the case being investigated by another agency, etc. Sanctions for non-compliance may be warnings (verbal or written), simple caution or prosecution. As prosecution is a costly and time-consuming process, emphasis is on warnings or simple cautions.

Types of frauds detected by the VSEB are essentially of three categories:

- 1) deliberate infection;
- 2) substitution of reactors;
- 3) inflating the value of the herd.

From 1st January 2007 to 24th November 2011 the closed files for Brucellosis offences have been 102, the majority of them related to failure to present for testing. DNA analysis is also used to help investigators in proving substitution of reactors or their progeny, as well as to verify pedigree claims.

7. Management of the Brucellosis Programme

Owen Denny

Mr Denny, SPVO of the Enzootics Section with responsibility for implementation of the Brucellosis Programme, explained how the current BB programme is staffed, organised and implemented. He began by outlining the structure at the top level in the Department of Agriculture and the geographical relationship of the 10 Divisional Veterinary Offices in NI and the DVO Sub-areas. He then reviewed the strategic response by the Veterinary Service to Brucellosis. The measures employed were outlined.

The staffing of the 10 veterinary offices was reviewed and numbers of vets and support staff outlined. The delivery of Veterinary Service Work was precisely

analysed and reviewed. The planning was broken down on a semester bases. Objectives are fixed and evaluated monthly for DVO.

The range of reports available for the management of Brucellosis was then outlined:

- Are tests allocated correctly?
- Are herd keepers notified of due risk tests?
- Are tests arranged on time?
- Are restrictions applied for overdue tests correctly?
- Are enforcement procedures in place for o/d tests?
- Are samples processed within target time?
- Are tests being checked in lab on time?
- Are queries at the laboratory getting sorted out on time?
- Are VO's interpreting/following up tests on time?
- Correct interpretation?
- Are restrictions applied correctly
- Are admin staff progressing tests?
- Are valuations being completed on time?
- Are valuations processed by admin within target time?
- Are valued reactors/NIC's slaughtered within target times?
- Brucellosis breakdown follow up tasks: VO visit, traces, mapping, Cleansing and disinfection complete, Segregation notices, CII complete, Investigation report

The detail of the Brucellosis audit was then presented. This consists of an Excel spreadsheet with 51 questions that are asked during the audit of the DVO. The Quality Control of the BB programme was illustrated in detail. The vision of eradicating brucellosis by March 2013 was presented.

8. Performance Monitoring, APHIS and Brucellosis Manager's Toolkit

David Brown

The Computer Database according to the Council Directive 64/432/EEC and Regulation 1760/2000/EC provides information about the animal's tag number, DOB, sex, breed and color, tag number of the dam, full movement history and DOD. Regarding holding, it should provide information about herd number, herd keeper details (name and address) and list of the animals present at time.

APHIS (Animal Public Health Information System) is a computer database system used by the Veterinary Service which provides information about animal traceability (animal details and movement history), animal health information (enzootic and epizootic – facilitating trade) and public health information concerning milk, meat and exports.

All livestock-related businesses are registered as enterprises – farms, markets, abattoirs, PVPs, EACs, shows, hauliers – and, within farm enterprises, bovine/goat/pig herds & sheep flocks. All bovine animals are registered – in the natal herd or first post-import herd. Tests and inspections can be allocated at enterprise, herd and individual animal level. All users and testing officers are registered.

DARD staffs are direct users of APHIS and record information such as blood sampling data, BB test results, BB test interpretation and further test allocation directly on APHIS. Operational partners such as PVPS, AVIs, markets, meat plants add information via the internet (TB tests, BB pre-movement test, exports, record animal movement in, out and deaths). Herd keepers and agents (tag suppliers and manufacturers) provide information via the internet. Herd keepers record animal births, movements and deaths to APHIS-on-line (AOL).

Herd keepers notify births by MC1 form, phone or electronically; colour, breed, sex, DOB, dam identity (sire identity) are also registered on the

system. All moves are recorded from herd keeper notifications and from market/abattoir inputs, including holding numbers & dates of movement. Deaths are recorded from herd keeper notifications and abattoir/rendering plant input, including date of death & means of disposal.

Information regarding bovine herds includes:

- Herd keeper details – name, address, map reference;
- List of animals;
- Testing review and preview;
- Movements to & from herd;
- Tags issued and not yet used;
- Movement notification documents issued and not yet used;
- Associated herds (epidemiological links);
- Out farms;
- Contiguous herds.

Allocated tests are performed at herd or individual level and the following are recorded: due date of test and to whom the test is allocated (patch/area for DARD test or PVP practice code). They are classified by category and type progressed in a cycle, each stage described by a status. Each stage of the test cycle is recorded by APHIS as well as the user code of the person and time of action; all are fully auditable.

APHIS traceability includes forward and backward tracing and allows the user to build an historic herd list and a list of adjacent herds.

The BB management & manager's toolkit seeks to:

- Identify key stages in test 'cycle';
- Set targets for each stage;
- Measure performance against targets using Cognos reports;
- Produce regular performance reports;
- Identify issues – e.g.: overdue tests, incorrect decisions;
- Take action – training, reallocate resources.

The range of reports available for the management of Brucellosis was then outlined, which provides the following checks:

- Are tests allocated correctly?
- Are herd keepers notified correctly?
- Are tests arranged on time?
- Are tests progressed correctly?
- Are we using enforcement procedures?
- Are test results interpreted correctly?
- Are admin follow up actions taken on time?
- Are reactors valued/ slaughtered on time?
- Are breakdown procedures correct?

The targets are set as follows:

- Sampling:
 - Post-abortion tests – 7 days after due date;
 - Risk herd tests – 30 days;
 - Individual tests – 30 days;
 - Routine tests – 60 days.
- Dispatch of samples – 3 working days after sampling
- Check in at Lab – 5 working days after sampling
- Total test cycle (sampling to interpretation) – 9 days
- Interpretation of positive tests – 24 hours
- Reactor removal – 15 working days after sampling

Monitoring performance and toolkit reports

Overdue tests:

- CA1s & CA2s due more than 7 days;
- BR risk tests 30 days or more overdue;
- Individual Br animal tests overdue 30 days or more;
- Routine BR herd tests overdue 60 days or more.

Test Progression:

- BR tests at Confirm 5 or more days;
- BR tests at Samples more than 7 days.

Enforcement – Restriction of herds

Correct interpretation

Valuation tests now due

Animals valued but not yet dead

Measuring of the performance by Matrix reports which are produced every month and divided into KPIs and PIs

Testing performance:

- Records performance in previous month
 - Red < 75 %
 - Amber: 75 – 84 %
 - Green >= 85 %

Breakdown management:

- Records performance in month previous to last
 - Red < 80 %
 - Amber: 80 – 89 %
 - Green >= 90 %

Sheep and goat sampling started in 1/11/2008. Six flocks were sampled, confirmed BB BR1 premises, one flock from a sheep flock on 'At-risk' premises and seven feral goats in a high risk area. All samples were tested with RBT using *Brucella abortus* antigen and all results were negative.

9. Field DVO Brucellosis Management

Nigel Trimble

The organisation of the Newry divisional area and implementation of the brucellosis programme in the region were presented. The monitoring

procedures include annual herd testing of all herds while inconclusive reactors must be retested. Cows with abortions or stillbirths are immediately tested and retested after 21 days. Bulk milk ELISA, abattoir sampling and pre-movement testing are some other monitoring procedures.

Field VOs cover a geographic area within the DVO. AHWIs are assigned to sub-areas of these patches and are responsible for keeping testing up to date while SAHWI and Group 4 manager monitor overdue tests.

A series of timely checks is used to:

- Monitor high risk tests and breakdown procedures;
- Highlight overdue tests;
- Highlight tests with problems, e.g. cancellations or samples missing;
- Monitor routine tests to organize resource and check allocations;
- Instigate legal action for failure to test.

Test results are reported on APHIS by the staff at AFBI. Interpretation is either automatic or manual and is interpreted as per staff instructions. Follow-up tests and restrictions are assigned. All paperwork is generated by administrative support staff.

Abortions are reported on farm, by a phone call or online through the Rural Portal. A daily email list is submitted to AFBI Laboratory. Stillbirths are notified by post. Details are recorded and entered on a spreadsheet while animals are restricted and test is allocated. Further tests are allocated for 3 weeks at initial test interpretation.

In the case of a breakdown, the Field VO is responsible for interpretation of reactor test results, application of restrictions, setting of tests (APHIS), and liaison with all those involved in the breakdown e.g. the farmer, private veterinarian and health and safety executive. An initial map is created, an outbreak file opened and the various databases updated. At the breakdown visit, the VO discusses the notices issued to the herd owner (e.g. herd

restriction notice, cleansing and disinfection notice and segregation notice), completes the investigation report and initial mapping and provides information about public health aspects. Cleansing and disinfection of the farm premises, isolation/segregation are checked by the VO. Animals are valued and slaughtered while samples are sent to the AFBI Laboratory. Culture results and further serological results must be reported on APHIS. If infection is confirmed, the herd may be depopulated. Further cleansing and disinfection of the premises must be done, while the slurry must be treated with lime. Restocking tests are conducted prior to the OBF status being restored. Risk testing in the area, based on epidemiological factors, is undertaken. Serological testing of sheep on the farm premises or in the area is undertaken if possible. Monitoring of the field procedures is achieved by the Brucellosis Management Team.

The brucellosis database and how the different tools work was then presented:

- Breakdown management tool;
- Work recording tool;
- Overdue test management tool.

The presentation concluded with the history of BB in Newry DVO. Information of annual BB herd incidence in other DVO's of NI was given. In 2010 there were 12 confirmed outbreaks while in 2011 there were 3. Reactor animals were 56 in 2009, 58 in 2010 and 9 to the end of September of 2011. An incident in Lislea (Armagh) was also mentioned, where an apparent attempt to infect a cattle herd with brucellosis, by dumping an infected fetus in a field, was described.

10. Local management of Brucellosis breakdowns - Armagh DVO

David Kyle & Elaine Dickson

Armagh is located in the south of NI and borders the Republic of Ireland (Counties Monaghan and Louth). There are 3044 herds of which 754 are

dairy, 378 LFA cattle and sheep and 1912 Lowland cattle and sheep. The number of staff at the Armagh DVO includes 7 VOs, 1 Enforcement Officer and a SAHWI, 19 AHWI's and 4 Administrative Staff.

Economic, social and political factors influence BB eradication in Armagh area. Most of the farms consist of small holdings and there is a decline in agricultural incomes. There is a culture of cattle trading and difficulties in enforcing regulations. Another influential factor is the border with the Republic of Ireland, with vehicular and people movement across the border. A graph with the new herd breakdowns that were confirmed by bacteriological culture was then presented.

Elaine Dickson described the brucellosis cluster of breakdowns in Keady which started in January 2010 and resulted in additional control measures in Armagh DVO. There was an increase in the frequency of risk testing (inner ring [IR] and outer ring [OR]) from every 3-4 months to every 2 months and an increase in the number of risk tests required before de-restriction of inner and outer ring herds (IR – 3 tests v 2 tests OR – 2 tests v 1 test) A small temporary control area was formed to prevent spread of the disease outside the area and to prevent 'clean' herds being brought into the area. These measures increased awareness and improved the rate of reporting abortions. The tracing of Inner Ring Herds was undertaken to detect any positive animals that may have left a herd before risk testing started (approx. 38 herds, 100 animals.) A large control area was then introduced, which included approx. 300 herds. This was necessary because there was a lack of cooperation from a herd keeper with a confirmed breakdown, and to provide extra assurance to ROI. In addition, information was received that a herd was planning a breakdown.

Abattoir surveillance was undertaken to ensure herds could not dispose of high risk cattle without testing. A marker was applied to herd on APHIS to notify inspectors in the abattoir. All older cows presented for slaughtered were blood sampled. From Aug 2010 to Jan 2011, 40 herds involved approx. 600 animals were highlighted in this way.

Bulk milk testing was increased. All sampling was carried out bi-monthly by AHWI's from high risk dairy herds and herds on the same milk collection as breakdown herds.

Herds with confirmed disease that were not depopulated (3) had the following measures applied at herd tests:

- Reduction in testing age to 6 months;
- Use of Rose Bengal Test;
- Strict interpretation.

There was an increase of Compliance Checks with inspections carried out every 2 weeks by AHWI's during the summer grazing of 2011, to ensure compliance with segregation notices (breakdown herds and inner ring herds).

The source of the disease for the Keady cluster remains unknown. It was thought that illegal cattle movement and/or high throughput of high risk animals may have introduced the infection, with some natural spread within the cluster.

The implications of the temporary control area (TCA) were that all herds were restricted in the extended area and there was no movement until the testing was completed. There was an increase in high risk tests and bio security visits were more intensive.

The benefit of the small TCA was that it controlled movement and disease spread while the large TCA did not control movement but did control disease spread. Both TCAs increased public awareness and served as a vehicle for public opinion, with farmers' meetings held in June and September 2010. The TCAs also demonstrated farmer compliance and commitment by the Veterinary Service.

There were 3 breakdowns in Armagh in 2011, one of which was confirmed, with the source suspected as being mechanical transfer. So far there has been no contiguous spread of the disease and no unexplained outbreaks.

11. Brucellosis serology

Alastair Douglas

Brucellosis serology

Since the last visit of the expert group in 2008, the number of samples tested by VSD has decreased a little bit after the previous increase due to the introduction of pre-movement testing in 2004 and introduction of parallel testing by both SAT and I-ELISA in 2006 (Fig.3).

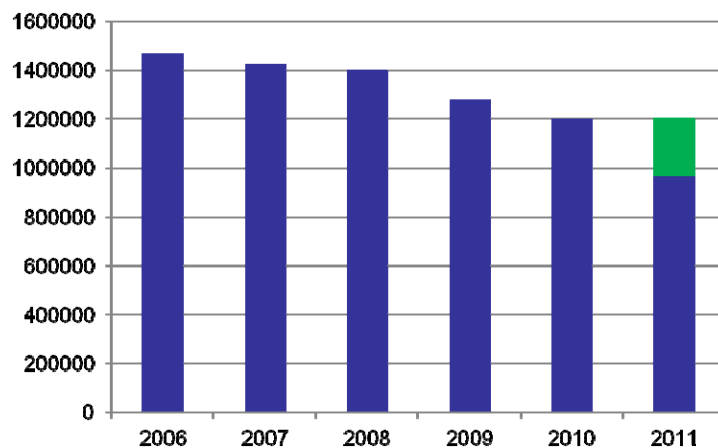


Fig.3: Serum samples tested per year at the VSD

The breakdown of samples types per year is: (i) Field eradication (ca. 950,000 samples), (ii) pre-movement testing (ca. 170,000), (iii) abattoir (ca. 25,000) and (iv) bulk milk testing (ca. 30,000 samples) (Fig.4).

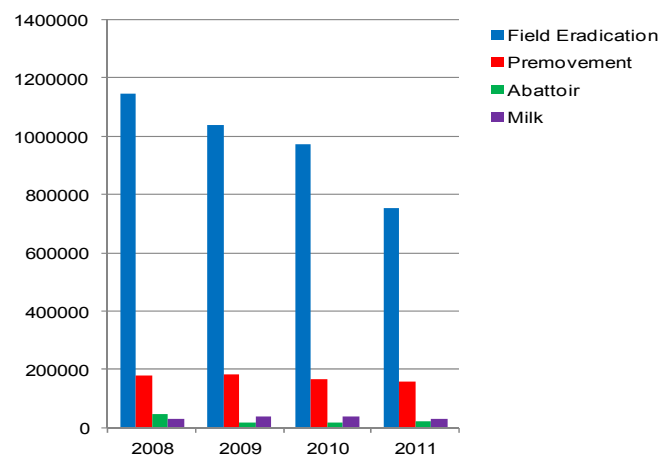


Fig.4: Breakdown of samples types

Submission rates rise through the winter, peak in spring and fall off again in summer (Fig. 4). The laboratory deals with this by (i) ensuring lab and equipment capacity for peak demand, (ii) imposing complementary seasonality on other work programmes and (iii) transfer of staff.

The tests currently performed are microtitre serum agglutination test (MSAT), MSAT with addition of EDTA, Complement fixation test and I-ELISA. The lab has also been performing C-ELISA and Rose Bengal test for specific studies, particularly the “Parallel trial” done in 2005. Antigens and kits are of commercial sources and comply with OIE requirements.

Serum samples are screened by MSAT (at 1:10, 1:20 and 1:40 dilutions) with a cut-off of 25 IU/ml.

The selection of non-negatives for further testing depends on the identified level of risk:

In low risk samples (Fig. 5), if samples are non-negative in MSAT at screening (≥ 25 IU), they are titrated to end-point by MSAT, EDTA-MSAT and CFT. For abattoir survey samples the EU cut-off of 30 IU/ml is used (Annex C of the Directive 64/432/EEC). From 2012, the test cut-off will be adjusted to 30 IU and non-negatives on the screening MSAT will be tested by I-ELISA, MSAT titration and CFT.

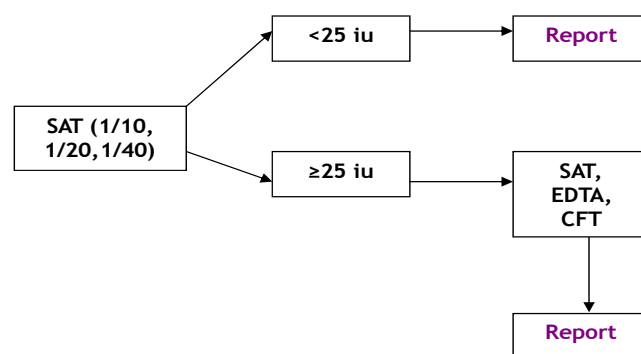


Fig.5

High risk samples (Fig. 6) are tested in parallel in both MSAT and I-ELISA. If either test is positive, sera are titrated to end-point by MSAT, EDTA-MSAT

and CFT. From 2012, the MSAT cut-off will be adjusted to 30 IU and non-negatives will be tested by MSAT titration and CFT.

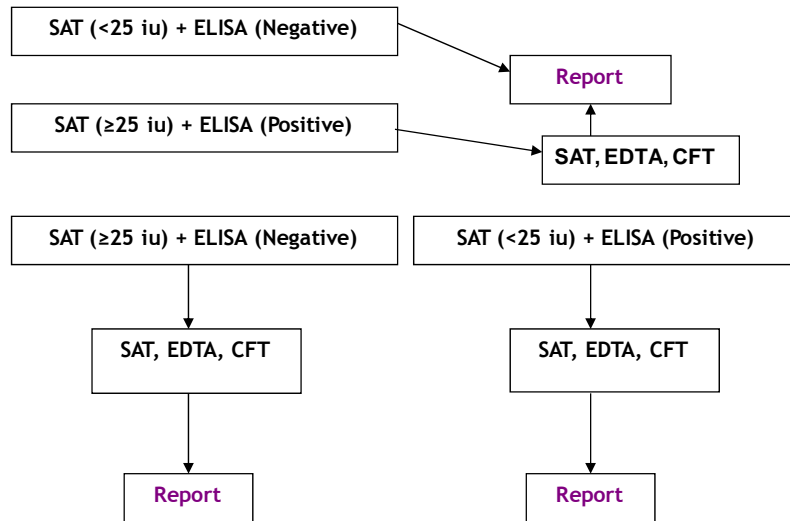


Fig.6

Numbers of tests carried out according to the year and tests categories are showed in figures 7-9.

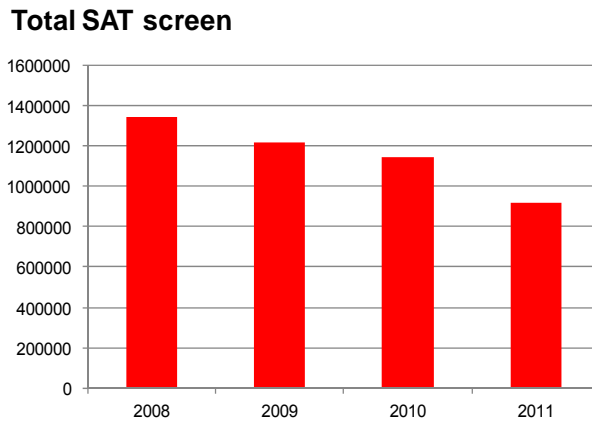
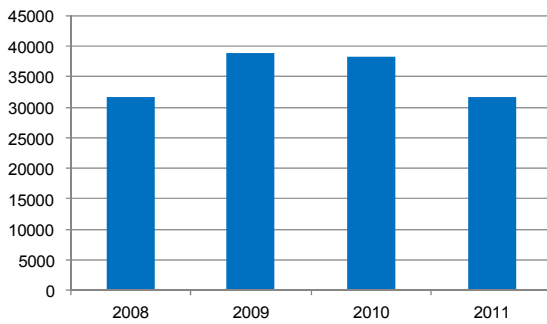


Fig.7

Total Milk ELISA



Total Blood ELISA

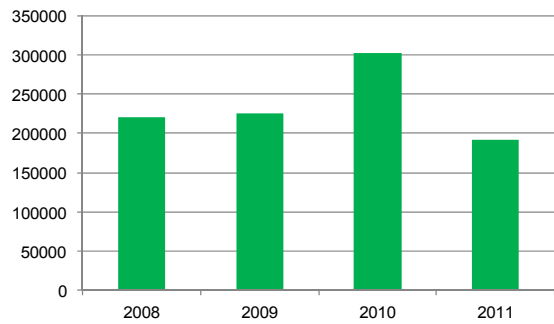


Fig.8

Total Confirmatory Tests

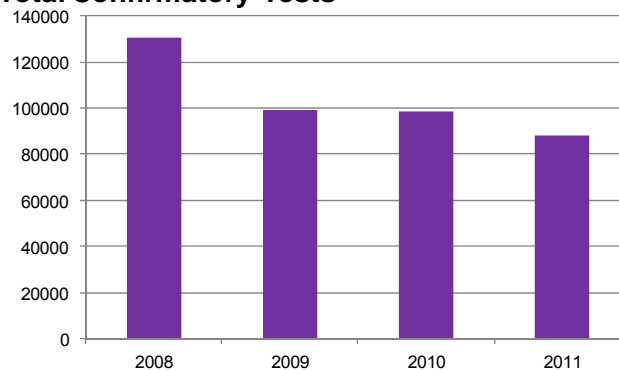


Fig.9

Information management and sample tracking of serum samples are as follows:

Each submission is accompanied by form BT20a where details of animals sampled are given and that also functions as lab work-sheet. Samples and details are checked and discrepancies are identified for according to a procedure. The negative results on screening are entered on BT20a and validated by lab manager. The non-negative sample details are transferred to lab work-list for further tests and final results are entered on BT20a and validated by lab manager. The results are then entered on APHIS and validated by work area manager. The hardcopy original BT20a is completed, and work-sheets and work-lists are archived.

Information management and sample tracking of milk samples are as follows:

Samples are received from dairies, bar-coded by producer number. The results are currently reported manually but an electronic transfer system is nearing completion.

The tests are carried out according to requirements laid out in Technical Annex of Directive 64/432/EEC.

The serology lab has been recently accredited according to ISO 17025 standard as required by Regulation 882/2004/EC, and participates regularly to the ring-trials organised by the EU-RL as well as by AHVLA VETQAS with all results within acceptable ranges.

The lab also includes daily strong and weak field positive in all tests as well as saline and negative controls in MSAT, RBT and confirmatory tests. Other quality assurance measures include: in MSAT and confirmatory tests, 1 % of samples chosen at random for repeatability checks, cross checks of 1 % samples to ensure ongoing competency of readers. For I-ELISA (serum and milk) control cards have been put in place to monitor the continuous trend of ODs and indexes of controls. "High negative" samples (≥ 80 % of the test cut-off) are tested in duplicate and a 25 % tolerance limit is applied based on the mean of the duplicates.

A structured training regime for staff is in place for all tests, including objective evidence and reassessments. Finally, all results are validated by the lab manager prior to reporting.

A Laboratory Information Management System (LIMS) has been in development for a number of years and is currently in use for handling milk ELISA testing. The final documented validation (to ISO17025 standard) is underway and full go live across all testing is anticipated in spring/summer 2012. It will enable full traceability of samples through lab assigned sample barcoding, a connection of instrumentation in the lab to LIMS, thereby improving traceability and cherry picking of retest samples and direct reporting of results to APHIS (as per current milk arrangements). A "plate map interface" will be in place for lab staff to enter results and highly customisable search engines will enable specific information to be extracted on demand.

12. *Brucella* culture, characterisation and typing

Sam Strain

The laboratory facilities dedicated to *Brucella* bacteriology are operated in a new (rebuilt in 2008) building including containment level 3 bacteriology laboratories, one of them being dedicated to *Brucella*. The VSD holds the Specified Animal Pathogens Order (SAPO) licence. The lab has been audited for accreditation according to ISO 17025 with a very favourable audit report.

Samples submitted to *Brucella* culture are collected at slaughter from high-titre animals (since 2007: parotid, retro-pharyngeal, sub-mandibular (head), supra-mammary in female [or superficial inguinal in males] and iliac lymph nodes) or from freshly aborted animals (vaginal swabs from aborted animals in high risk herds and fetuses and aborted material submitted by private veterinary).

The procedure used for culture is according to the OIE Manual and to the UK National Reference Method (NRM004).

For primary isolation from tissues, all 5 lymph nodes are cultured individually selective Farrell's medium (2 plates per lymph node), and incubated at 37°C in 10% CO₂ for 14 days (with a reading every 3-4 days).

As regards identification of species and biovars, the OIE manual recommendations are followed except for small details with several specific measures aiming at ensuring a high level of both bio safety and traceability.

All isolates in Northern Ireland since 2008 have been characterised as typical CO₂-dependent *B. abortus* biovar 1 strains.

Figure 10 shows the number of animals sampled and the number of samples submitted to *Brucella* culture at VSD in comparison with numbers of reactors at NI level (1995-2011).

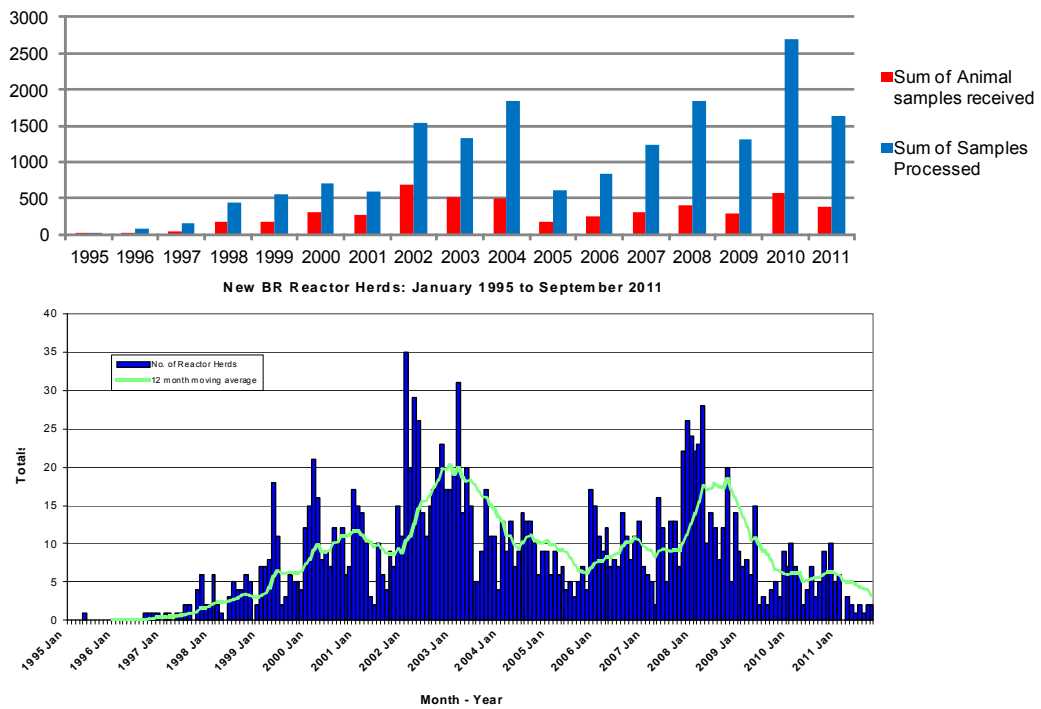


Fig.10

Figure 11 gives the overall percentage of samples testing positive in culture.

Percentage samples testing positive

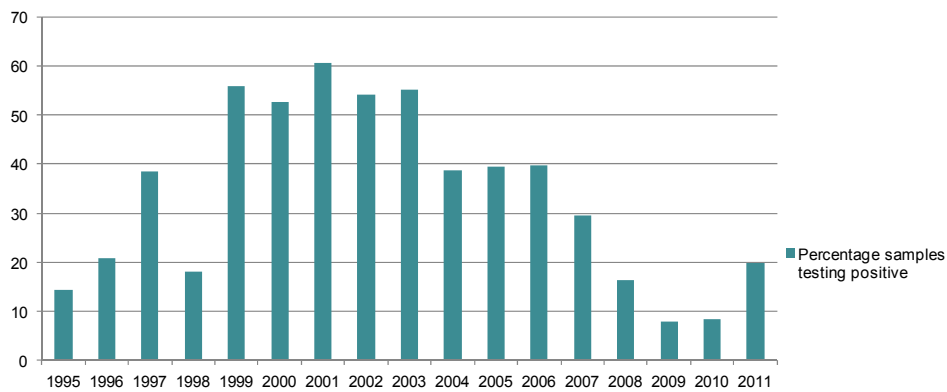


Fig. 11

Figures 12-15 give details on respective isolation rates according to the kind and numbers of lymph nodes cultured. Results are in accordance with those reported elsewhere showing that the supra-mammary LN gives the highest

rate of isolation and that the culture of at least 4 different LN optimises the global isolation rate.

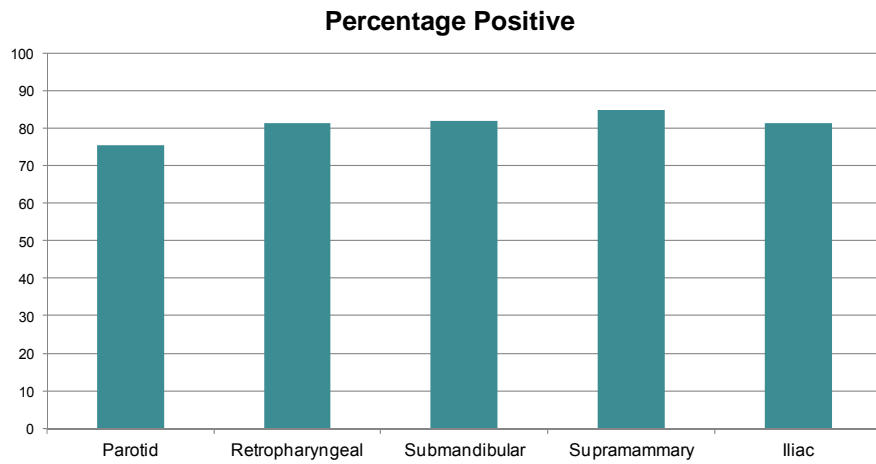


Fig.12: Isolation rates for individual lymph nodes from culture positive animals (2007-2011)

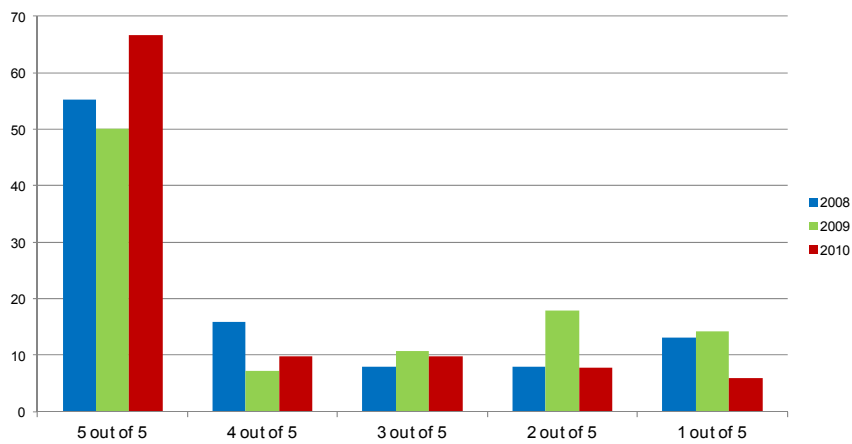


Fig.13: Percentage of multiple positive tissues from culture positive animals (2007-2011)

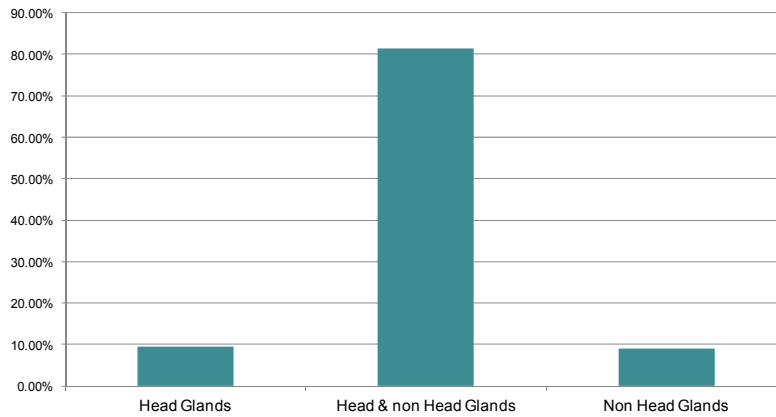


Fig.14: *Brucella* isolation from different body systems (2007-2011)

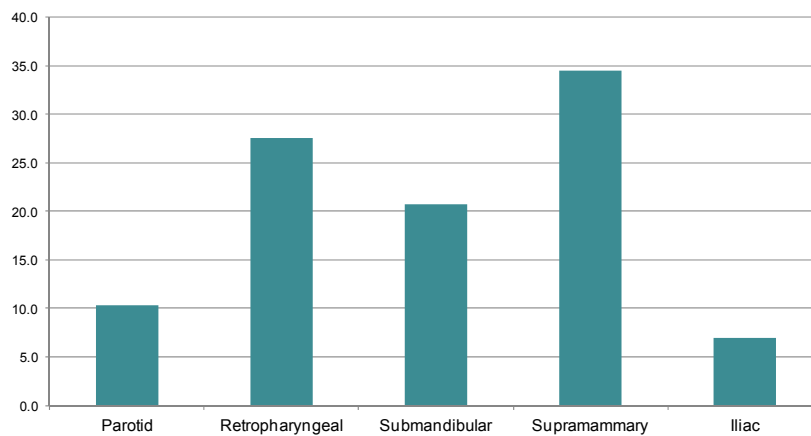


Fig.15: Percentage of culture-positives in one tissue site (2007-2011)

13. Host and pathogen genotyping

Robin SKUCE (Genotyping)

At VSD, molecular biology is used for assisting the bovine brucellosis eradication programme in both animals and *Brucella* genotyping.

As far as animal genotyping is concerned, the methodology is based on the short tandem repeat (STR) polymorphism analysis (11 STRs for identification and on 17 STRs for parentage). This allows verification of the proposed identity or parentage. For identity, there is a risk of error of $1/590 \times 10^6$ (probability that 2 animals with the same identity) and for parentage, of $1/262 \times 10^3$ (probability that a sire or a dam produce another calf of the same identity). The test is applied to:

- To verify and audit queried animals (cattle identity, parentage, pedigree);
- To detect and deter fraud;
- In case of late calf registration;
- For biopsy-tag bovine TB reactors and OTMS (cattle over 30 months for human consumption) abattoir audit.

More than 750 animals have been tested in 2010 and a biobank of suspect animals has been set up. The method and its application in VSD were accepted by UKAS and by an external technical expert.

As far as *Brucella* genotyping is concerned, the MLVA (VNTR analysis) has been assessed by VSD as a potential tool for giving additional epidemiological information as regards the relationship between breakdowns at historical and geographical levels on 926 isolates from 372 herds (2000 & 2004-2011).

The results disclosed within-herd multiple isolates and within-animal multiple isolates showed that some markers do not discriminate this sample at all. Some markers showed unacceptable instability. Multi-locus assay selects markers which better balance discrimination and stability.

Some profiles were identified with a marked degree of geographic localisation. This was not evident with 26 VNTRs or Hoof-Prints alone. However results could be consistent with epidemiologically-linked outbreaks?

Therefore VNTR 'genetics' allows relationships between strains to be inferred. As a conclusion of this study, the markers and assay merit further investigation. However, it is perhaps too late for NI since the control in place has progressively purged the strain diversity. New methodologies have to be investigated in order to help in confirming or excluding the sources of infection. Herd situation at present is 150 bovines (90 dairy).

DAY 2

1. Farm visit

A farm was visited in the morning of day 2. The holding was located close to Mullaghbane in South Armagh, about 10 miles from Newry and 3-4 miles from the border to the Republic of Ireland.

The holding was a mixed herd with 170 bovines, mostly dairy with some fattening. From October to March, animals are housed on the farm, while grazed on pastures near the farm during the remaining period. Vaccination against BVD and IBR is regularly applied. Holding was BB free for nearly 10 years. The herd was considered at risk because in 2010 a few outbreaks were registered in the area. Testing was therefore carried out with a frequency of less than 6 months. In December 2009 a BB infected foetus was found near the premises and the owner suspected a deliberate action by an unknown party to infect the area and the herd.

In January 2010 the entire herd tested negative to BB. During the months of March 2010 a total of 8 abortions were found in the herd. A total of 14 animals were found positive at a first testing and 2 more positive animals were found at a second testing. Because of these findings the herd was depopulated on April 10th 2010. After cleaning and disinfection the holding was restocked 6 months later. The restocking was achieved by buying bovines from various different farms.

CONCLUSIONS

The Bovine Brucellosis Task Force Subgroup recognises the significant efforts made by the NI Central Authorities and the substantial progress towards eradication of brucellosis. The group was particularly pleased to observe (1) the level of organisation, including the use of a dedicated management group with shared goals and strategies, operating from central to local level and (2) the amount and quality of data collected and used to manage the programme. The Task Force was also pleased to see improvements made both in the programme and in the laboratory; almost all recommendations made by the subgroup in 2008 have been implemented.

The group noted however, that although the system of compensation has been reviewed, no changes have been implemented and no upper limit has been set as previously recommended. This may provide an important obstacle to achieve eradication.

RECOMMENDATIONS

1. Continue with the measures being employed, especially in the higher risk areas. Reduction of measures might be considered in free areas but current levels of surveillance should be rigorously maintained in the rest of the country. In higher risk areas, implementation of additional measures (e.g. testing of young animals, extended use of brucellin skin test, and use of parallel testing) might be considered but their impact on the sensitivity and specificity of the surveillance system should be assessed beforehand.
2. Since depopulation remains the best method to prevent latent carriers, careful consideration should continue to be given to any herd that is not depopulated.
3. An upper limit (cap) for compensation should be set. The compensation levels and systems in other MS should be taken into account to learn from their experience.

**MEETING OF THE BOVINE BRUCELLOSIS SUB-GROUP OF
THE TASK FORCE FOR MONITORING DISEASE ERADICATION
HELD IN NEWRY, NORTHERN IRELAND, UK, 06-07 DECEMBER 2011**

PARTICIPANTS

Task Force Sub-Group – members

- Ernst STIFTER, Chairman, Italy
- Bruno GARIN-BASTUJI France
- Fabrizio DEMASSIS Italy
- Michalis COSTOULLOS Cyprus
- Darrell ABERNETHY UK (Northern-Ireland)

Excused: Olga Minguez (ES), Antonio Pina Fonseca (PT).

Northern-Ireland Representatives (main list)

▪ Owen DENNY	Senior Principal Veterinary Officer (SPVO) of the Enzootics Section
▪ David BROWN	Enniskillen DVO
▪ Sharon VERNER	DVO Brucellosis, Enzootics Section
▪ Ian McKEE	Grade 7, TB/BR Policy Branch
▪ David KYLE	Armagh DVO
▪ Elaine DICKSON	Armagh DVO
▪ Nigel TRIMBLE	Newry DVO
▪ Danny GRAY	Deputy Principal Enforcement Officer
▪ Judith GRAHAM	VO – Brucellosis section
▪ Alastair DOUGLAS	AFBI, VSD, Immunodiagnostic Branch
▪ Samuel STRAIN	AFBI, VSD, Bacteriology Branch
▪ Robin SKUCE	AFBI, VSD, Bacteriology Branch
▪ Darrell ABERNETHY	Epidemiology DVO and Task Force member

Annex II

MEETING OF THE BOVINE BRUCELLOSIS SUB-GROUP OF THE TASK FORCE FOR MONITORING DISEASE ERADICATION HELD IN NEWRY, NORTHERN IRELAND, UK, 06-07 DECEMBER 2011

AGENDA

Timing	Item	Speakers
DAY ONE	NEWRY Divisional Veterinary Office (DVO)	
09.30	Introduction and Overview	Owen Denny
10.00	Epidemiology – the current picture in NI	Darrell Abernethy
11.00	<i>Tea/Coffee</i>	
11.15	Northern Ireland Brucellosis Scheme	Sharon Verner
12.00	Compensation update	Ian McKee
12.15	<i>Questions and discussion</i>	Owen Denny
12.30	<i>Lunch</i>	
13.15	BR Programme Management	Owen Denny
13.45	Local management of BR breakdowns	Nigel Trimble
14.15	HQ management of BR breakdowns	Judith Graham
14.45	<i>Questions and discussion</i>	
15.00	<i>Tea/Coffee</i>	
	Agri-Food and Biosciences Institute (AFBI) procedures	
15.15	<i>Brucella</i> serology	Alastair Douglas
15.45	<i>Brucella</i> culture, characterisation and typing	Samuel Strain (TBC)
16.15	Molecular typing of <i>Brucella abortus</i>	Robin Skuce
16.45	<i>Questions and discussion</i>	
17.00	close	
DAY TWO	NEWRY Divisional Veterinary Office (DVO)	
09.30	Farm visit	
	<i>Return to NEWRY DVO</i>	
11.30	<i>Tea/Coffee</i> and Final opportunity for questions	All Speakers
12.00	EU Task Force BR subgroup meeting	
13.00	<i>Lunch</i>	
13.45	Final conclusions and recommendations	TF Subgroup
14.30	close <i>Tea/Coffee</i>	