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Introduction

This is the third Annual Report on the animal disease surveillance activities of the Department of Agriculture, Fisheries and Food Regional Veterinary Laboratory Service. The report provides a representative summary of diseases detected in farm animals – either in carcasses or clinical pathology material submitted to the laboratory, or as a result of on-farm investigations by laboratory staff.

The health status of Irish livestock is of great importance to the national economy. Approximately ninety percent of our agricultural exports are animal based. Livestock production alone contributes in excess of €4.1 billion to the Irish economy annually. The Regional Veterinary Laboratories (RVLS) form a key component of the State’s surveillance of statutorily-controlled endemic and exotic diseases, e.g. tuberculosis, brucellosis, BSE, scrapie, Avian Influenza, Foot and Mouth Disease etc. A vigilant laboratory surveillance network contributes to maintaining the health of the national herd by providing up-to-date information on the occurrence of endemic diseases, and by the early identification of new or exotic diseases. Recent outbreaks of such exotic diseases in other jurisdictions – e.g. Foot and Mouth Disease, Avian Influenza and Bluetongue in the UK – are a timely reminder of the importance of the surveillance role of the laboratory network, not alone to the agricultural industry, but to the economy as a whole.

The Regional Veterinary Laboratory network, established in 1967, forms an integral part of the national animal disease surveillance programme. The RVLS are strategically placed in Sligo, Athlone, Dublin, Kilkenny, Limerick and Cork to provide optimal coverage of the surrounding livestock populations. They provide a diagnostic pathology, investigative and advisory service to the agriculture industry. Clinical samples from live animals, or carcasses of dead animals, are submitted to the RVLS on a daily basis. The diagnostic investigations conducted by veterinary pathologists and laboratory analysts seek to aid diagnosis, facilitate treatment, and promote disease prevention, thereby optimising herd health and animal welfare.

Compilation of this report has been facilitated by analysis of data from the laboratory service Laboratory Information Management System (LIMS). This has been in operation throughout the laboratory service since 2003. It is an integrated system which links the regional and central laboratories in real time, and records all submission information from sample receipt, through testing, to authorisation and final reporting. It provides a central database on which details relevant to individual animal disease investigations can be recorded. Interrogation of this database facilitates easy and efficient access to information regarding disease investigation information across the RVL network.

It is envisaged that this report will be of benefit to herd owners, private veterinary practitioners, industry, researchers and farm advisors. It is important to acknowledge that much of the information contained herein is the result of passive rather than active surveillance. Nevertheless, the RVLS provide a unique and important source of data on the causes of illness and deaths in Irish livestock.
### Diseases of Cattle

#### Neonatal Calves (birth to one month of age)

Enteric infections and septicaemia/bacteraemia were the two most frequently diagnosed fatal conditions in neonatal calves up to one month of age (24 per cent and 22 per cent respectively of total diagnoses). Rotavirus was the most commonly detected enteric pathogen (see Bovine Neonatal Enteritis, page 5). *E. coli* (45.8 per cent) and *Salmonella dublin* (13.5 per cent) were the most frequently isolated pathogens from calves diagnosed to have died from septicaemia/bacteraemia.

#### Calves (one to three months of age)

In calves one to three months of age, respiratory infections, at 27 per cent, were the most frequent diagnosis, followed by enteric infections (13 per cent), and septicaemia/bacteraemia (11 per cent). The agents most frequently identified in cases of respiratory infections were *Mannheimia haemolytica* (11.5 per cent), *Pasteurella multocida* (8.9 per cent) and *Arcanobacterium pyogenes* (7.7 per cent) – a pattern similar to that in the calves under one month of age.

Coccidia species were the pathogens most-frequently identified in calves with enteric infections (24 per cent). *Salmonella dublin* (39.4 per cent) was the pathogen most frequently associated with septicaemia/bacteraemia diagnoses, followed by *E. coli* (12.1 per cent). Diagnoses grouped under the heading ‘Gastro-intestinal obstruction/torsion’ (8 per cent) mainly comprise conditions such as mesenteric, gastric and intestinal torsions. The category ‘Nutritional/Metabolic Conditions’ includes ruminal and metabolic acidosis, bloat, dehydration and copper deficiency.
Weanlings (three months to one year)

At 30 per cent, respiratory infections topped the list of diagnosed causes of death in weanlings from three months to one year of age. This is comparable to the figure reported for 2006. Of the animals diagnosed with a respiratory infection, the bacteria Pasteurella multocida (22.3 per cent), Mannheimia haemolytica (16.1 per cent), together with the lungworm Dictyocaulus species (8.9 per cent), were the most frequently detected pathogenic agents. Enteric infections, at 7.5 per cent, were responsible for a smaller proportion of deaths than in the younger age groups.

Poisonings, and BVD virus infection, were also significant diagnoses as causes of death in this age group. Acute haemorrhages in the abomasum, uterus, spleen or brain were also recorded as causes of deaths. Nutritional or metabolic causes of death included hypomagnesaemic tetany, ruminal acidosis and fatty liver.

Clostridial Diseases in Cattle

Table 1 shows a breakdown by age of the types of clostridial disease diagnosed in cattle. Blackleg was the most common form in animals under 1 year of age. Four of these cases were in calves under 3 months of age. Clostridium sordellii was the organism most commonly associated with emphysematous abomasitis. Botulism and tetanus were only diagnosed in older animals.

Clostridial diseases are usually rapidly fatal and in severe outbreaks, many animals may die suddenly. Vaccination with a multivalent clostridial vaccine is a safe and reliable strategy for preventing clostridial disease.
Table 1: Age related incidence of Clostridial diseases in cattle 2007.

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>&lt; 1 year</th>
<th>&gt;= 1 year</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blackleg</td>
<td>38</td>
<td>1</td>
<td>39</td>
</tr>
<tr>
<td>Clostridial enterotoxaemia</td>
<td>12</td>
<td>2</td>
<td>14</td>
</tr>
<tr>
<td>Abomasitis- emphysematous</td>
<td>8</td>
<td>1</td>
<td>9</td>
</tr>
<tr>
<td>Botulism</td>
<td>0</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Black Disease</td>
<td>1</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Malignant Oedema</td>
<td>1</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Abomasitis</td>
<td>2</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Tetanus</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td><strong>Totals</strong></td>
<td><strong>62</strong></td>
<td><strong>12</strong></td>
<td><strong>74</strong></td>
</tr>
</tbody>
</table>

Table 2: Age related incidence of poisoning cases in cattle 2007.

<table>
<thead>
<tr>
<th>Agent</th>
<th>&lt;=1 year</th>
<th>&gt; 1 year</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lead</td>
<td>18</td>
<td>7</td>
<td>25</td>
</tr>
<tr>
<td>Ragwort</td>
<td>1</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>Fern</td>
<td>0</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Yew Tree</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Copper</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Rodenticide</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Unidentified</td>
<td>2</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td><strong>Totals</strong></td>
<td><strong>22</strong></td>
<td><strong>19</strong></td>
<td><strong>41</strong></td>
</tr>
</tbody>
</table>

Poisonings in Cattle

Table 2 shows a breakdown by age of the types of poisonings diagnosed during 2007. In the overall context, poisoning diagnoses, at 2.5 percent, represent a small percentage of the total. However, the inadvertent ingestion of lead still kills a significant number of animals each year.

Bovine Neonatal Enteritis

Enteritis continues to be the most common cause of mortality in calves less than one month of age (see Figure 1). The calf enteritis package is a series of tests performed by the Regional Veterinary Laboratories on submitted faecal samples to identify the causative pathogen of enteritis in calves less than one month of age. In order to facilitate identification of the specific pathogen involved in cases of calf diarrhoea, it is important that samples are submitted from recently infected animals – and preferably prior to treatment.

The relative frequencies of pathogens identified in faecal or intestinal content samples submitted in 2007 are shown in Table 3. Rotavirus was the most common at 28.3 percent. Cryptosporidium species were detected in 21.7 percent of samples tested, which represents a decline relative to 2006 (25.9 percent). K99 E. coli (1.1 percent positive) and Coronavirus (1.7 percent positive) were less frequently identified. Overall, the pattern was similar to that of previous years.

Table 3: Pathogenic agents detected in faecal samples submitted to the six RVLs for calf enteritis tests in 2007.

<table>
<thead>
<tr>
<th>Pathogen</th>
<th>Number Tested</th>
<th>Number Positive</th>
<th>Percentage Positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rotavirus</td>
<td>2561</td>
<td>726</td>
<td>28.3%</td>
</tr>
<tr>
<td>Cryptosporidium</td>
<td>2563</td>
<td>557</td>
<td>21.7%</td>
</tr>
<tr>
<td>Salmonella spp</td>
<td>2571</td>
<td>56</td>
<td>2.2%</td>
</tr>
<tr>
<td>Coronavirus</td>
<td>2561</td>
<td>44</td>
<td>1.7%</td>
</tr>
<tr>
<td>E.coli K99</td>
<td>1783</td>
<td>19</td>
<td>1.1%</td>
</tr>
</tbody>
</table>

The Zinc Sulphate Turbidity Test (ZST) was used to test a total of 684 calf samples in 2007 (Figure 5). Over a half of all samples tested (56 percent) had a value of less than 16 units - which is indicative of inadequate colostrum intake. A further 21 percent revealed values of 16 to 24 units - reflecting only moderate colostral intake. Only 23 percent of calves tested showed good colostral intake (i.e. greater than 24 units). These results serve to underline the role of good calf management in preventing calf mortality. This pattern was also evident among those calves tested for enteric pathogens; 57 percent of these were found to have inadequate colostrum intake.
intake (Figure 6). The proper feeding of colostrum, implementation of good hygiene, and the use of appropriate vaccination protocols, are necessary pillars in the prevention of calf enteritis.

Figure 6: Results of the zinc sulphate turbidity test indicating the level of colostrum intake in calves for which a sample had also been submitted for calf enteritis pathogen detection \( n = 450 \).

![Figure 6](image_url)

Figure 7: A positive Rotavirus fluorescent antibody test result on a calf faecal sample. (Photo: Martin Hill)

Coccidial infection is a significant cause of diarrhoea and/or dysentery in calves or cattle that are, for the most part, older than three months of age. The proportion of bovine faecal samples submitted for coccidial oocyst counts in which coccidial oocysts were detected has increased over the three years 2005 to 2007 (Figure 8).

**Figure 8:** Trends in the incidence of coccidiosis in calf faecal samples as a percentage of all faecal samples submitted to the RVLs for *Coccidia spp.* detection, 2005 to 2007.

Bovine Abortion

The Regional Veterinary Laboratories play an important role in the investigation of bovine abortions. Abortions represent a significant economic loss on farms. They may also pose a risk of human infection where agents such as *Brucella abortus*, *Listeria monocytogenes* and *Salmonella* spp. are involved. In 2007, the RVLs carried out a total of 1,647 bovine foetal cultures (foetal abdominal contents, foetal organs or placentas). The relative proportions of bacterial organisms isolated from these submissions follow a similar pattern to previous years. *Salmonella dublin* was the most commonly isolated microorganism in 2007. The table below compares values for the years 2006 and 2007:

**Table 4:** A comparison of bovine foetal culture results 2006 versus 2007.

<table>
<thead>
<tr>
<th>Agent</th>
<th>2006</th>
<th>2007</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Salmonella dublin</em></td>
<td>6.1%</td>
<td>7.0%</td>
</tr>
<tr>
<td><em>Arcanobacterium pyogenes</em></td>
<td>6.9%</td>
<td>5.8%</td>
</tr>
<tr>
<td><em>Bacillus licheniformis</em></td>
<td>4.1%</td>
<td>4.5%</td>
</tr>
<tr>
<td><em>Listeria monocytogenes</em></td>
<td>1.3%</td>
<td>1.6%</td>
</tr>
<tr>
<td><em>Aspergillus spp.</em></td>
<td>0.5%</td>
<td>0.9%</td>
</tr>
<tr>
<td><em>Brucella spp.</em></td>
<td>0%</td>
<td>0%</td>
</tr>
</tbody>
</table>
It is important to observe that, for a second consecutive year, *Brucella abortus* was not isolated in any of the samples submitted for examination in 2007.

**Figure 9: Salmonella dublin** positive abortions as a percentage of all foetal submissions during 2007. (n = 1,647).

*Salmonella dublin* has a well-documented seasonal distribution - with a peak in cases seen late in the year. In 2007, a rise in *S. dublin* cases was observed from July to November (Figure 9). The greatest number of cases was in November, at 37, representing 18.2 percent of the foetal submissions to the RVLS for that month.

*Salmonella typhimurium* (2 cases) and *Salmonella enteriditis* (1 case) were also isolated from abortion submissions in 2007.

Other microorganisms isolated from foetal material, but not listed in the above table, were *Escherichia coli* (102 cases, 6.2 per cent), *Streptococcus spp.* (26 cases, 1.6 per cent), *Staphylococcus aureus* (11 cases, 0.7 per cent), *Listeria spp.* (8 cases, 0.5 per cent), *Bacillus spp.* (7 cases, 0.4 per cent) and *Pasteurella multocida* (2 cases, 0.1 per cent). These organisms are sometimes isolated from healthy animals such that their presence in tissues of an aborted foetus cannot be considered as definitive evidence that they caused the abortion. An absolute determination of their significance may be assisted by analysis of associated data such as frequency of occurrence and the herd history.

**Table 5:** The frequency of other diagnoses associated with bovine abortion 2007.

<table>
<thead>
<tr>
<th>Aetiology</th>
<th>Number Positive</th>
<th>Number Tested</th>
<th>Percentage Positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>BVD virus</td>
<td>38</td>
<td>683</td>
<td>5.56</td>
</tr>
<tr>
<td>Neospora</td>
<td>38</td>
<td>771</td>
<td>5.22</td>
</tr>
<tr>
<td><em>L. hardjo</em></td>
<td>14</td>
<td>431</td>
<td>3.24</td>
</tr>
<tr>
<td>Dystocia / anoxia / hypoxia</td>
<td>50</td>
<td>1639</td>
<td>3.05</td>
</tr>
<tr>
<td>Hereditary / dev. anomalies</td>
<td>10</td>
<td>1639</td>
<td>0.61</td>
</tr>
<tr>
<td>Diagnosis not Reached</td>
<td>1011</td>
<td>1639</td>
<td>61.68</td>
</tr>
</tbody>
</table>

*Leptospira interrogans* serovar *hardjo*

*Leptospira interrogans serovar hardjo* is associated with milk drop syndrome and abortion in cattle and is of significant zoonotic potential, capable of causing human infection via infected foetal material or urine.

Foetuses and maternal blood samples are regularly submitted to the Regional Veterinary Laboratories for testing to detect *Leptospira hardjo* infection.

**Abortion is an important cause of production loss in the livestock industry. All on-farm abortions should be investigated, as effective control measures for abortions require prompt and accurate diagnosis.**

Autolysis (decomposition) of tissues reduces the sensitivity of the fluorescent antibody and foetal serology tests. In some cases, poor preservation may render a foetus completely unsuitable for laboratory examination. It is very important, therefore, to ensure delivery of aborted foetuses to the laboratory as soon as possible after they are discovered.
431 foetal carcasses were submitted for *L. hardjo* testing to the Regional Veterinary Laboratories in 2007, of which fourteen (3.2 per cent of submissions) were positive based on the presence of specific antibodies (Table 5). The fact that a foetus may be expelled up to 12 weeks after the dam has been infected is a complicating factor for the diagnosis of *Leptospira hardjo* abortion. Foetal antibodies may have declined to an undetectable level in the period between infection and the expulsion of the foetus.

Figure 10 presents the results of the solid phase immunoassay test for *Leptospira interrogans* serovar *hardjo* antibodies in 10,755 maternal blood samples, from animals of unknown vaccinal status, submitted to the Regional Laboratories in 2007. Positive results may indicate recent infection – or may be a response to vaccination. Interpretation of results can only be made based on knowledge of the herd history and vaccinal status of the animals.

**Bovine Mastitis**

3,658 bovine milk samples were submitted for bacterial culture and antibiotic sensitivity testing in 2007. This represents a 10 per cent increase when compared to 2006 - and possibly reflects the buoyancy experienced in the dairy industry during the year. 70 per cent of the samples submitted were from Munster counties.

Samples contaminated continued to be a problem in 2007. Contaminated samples typically yielded growths of coliform bacteria reflected in the high isolation rate of these. It is important that samples are collected aseptically into sterile containers, refrigerated immediately and delivered to the laboratory as soon as possible.
The seasonal pattern of sample submissions in 2007 was similar to previous years (Figure 12), with the numbers of submissions peaking in October and November.

*Staphylococcus aureus* was isolated from 35.3 per cent of the milk samples submitted to the RVLs, and was the most commonly isolated pathogen. Similar to the trends identified in previous years, the isolation rate varied by month, with the peak occurring in October (Figure 13).

**Figure 12:** Milk sample submission numbers by month: 2005-2007

Streptococcus agalactiae, a pathogen associated with serious herd outbreaks of mastitis, was a rare finding, being isolated from only 5 of the samples submitted (0.1 per cent).

**Figure 13:** Staphylococcus aureus isolation rates by month as a percentage of all submissions (n = 3,658).

**Bovine Respiratory Disease**

Bovine respiratory disease (BRD) is a major cause of illness and death in Irish cattle. It has a significant economic impact on the cattle industry because of increased mortality, treatment costs and decreased production (growth rates).

Figure 13 shows the number of PCR positive results recorded in CVRL for the four main viruses responsible for bovine respiratory disease between October 2006 and December 2007. As would be expected, bovine viral pneumonia is markedly seasonal, most notably for BRSV and to a lesser extent for IBR. The BVD figures reflect the roles of the BVD persistently infected animals, and of BVD co-infection with other viruses, in the pathogenesis of respiratory disease in Irish cattle. The Parainfluenza-3 PCR test was introduced in August 2007.

**Figure 14:** The number of PCR positive results recorded in the Central Veterinary Research Laboratory (CVRL) for the four main viruses responsible for bovine respiratory disease between October 2006 and December 2007.

Bacterial agents involved may include: *Mannheimia haemolytica, Pasteurella multocida, Histophilus somni* and *Mycoplasma bovis*. The incidences of each in pneumatic lungs examined post mortem in 2007 are indicated in Figure 15.
Johnes Disease in Cattle 2007

Johnes’s disease is a Class B notifiable disease. It is caused by infection with the bacterium *Mycobacterium avium* subsp. *paratuberculosis* (MAP). Cattle typically acquire infection as calves, either through ingestion of colostrum and milk containing MAP, or by exposure to MAP in faeces or contaminated feed and water. Clinical signs are slow to develop and are most-frequently seen in animals 2-6 years of age. The onset of clinical signs is influenced by a number of factors - including infection dose and production stresses. *In-utero* infection of the foetus can occur - and can be of significance in herds where the prevalence is 5 per cent or greater.

Over the three years 2005 to 2007, MAP was isolated from around 20 per cent of bovine faeces submitted to the RVLs for Johnes faecal culture (Table 6). Positive samples were from cattle of various ages - including heifers and older cows. A liquid culture system was introduced in the latter part of 2006. This has allowed culture results to be issued after 9 weeks, compared to the 16 weeks typically required for solid media.

The number of sera tested in the MAP ELISA has increased year on year. On average 7.3 per cent of the sera tested each year are positive. Where the possibility of Johnes’s disease infection is being investigated in a herd it is advisable to follow up seropositive animals with submission of faecal samples for culture which can help in identifying false positive serology results due to infection with non-pathogenic environmental mycobacteria.

As Johnes Disease is notifiable, it is important to submit the full tag number and herd number of each animal being tested.
Diseases of Sheep

Mortality in Lambs (birth to six months of age)

Figure 17 summarises the relative frequency of the diagnosed causes of deaths of 242 lambs under six months of age. The most commonly occurring diagnoses in this age group were septicaemia, enteritis, clostridial enterotoxaemia and pneumonia. Septicaemia was typically associated with *Mannheimia haemolytica* (older lambs) and *Escherichia coli* (neonatal lambs) while the most common form of clostridial enterotoxaemia in this age group was pulpy kidney disease (diagnosed in 18 of the 25 cases of clostridial enterotoxaemia). *Mannheimia haemolytica* was also isolated from the lungs of the majority of the pneumonia cases, which appeared to occur in two distinct peaks, in April-May, and in July-September. These patterns are broadly similar to those seen in previous years.

Diagnoses associated with the deaths of less than 1.5 per cent of lambs examined have been grouped together under “Other Diagnoses” and include conditions such as abomasal ulcers, anaemia, ruminal acidosis and louping ill.

Figure 17: The relative frequency of the diagnosed causes of deaths of lambs under six months of age (n = 242).

Mortality in Sheep (older than six months of age)

Figure 18 outlines the relative frequencies of the diagnosed causes of deaths of 222 adult sheep submitted for post mortem examination in 2007. Pneumonia, metabolic/nutritional diseases, and parasitic disease were the most common diagnoses in this group. *Mannheimia haemolytica* was the most commonly diagnosed agent associated with pneumonia in adult sheep. The heading ‘Metabolic/Nutritional diseases’ includes a range of diagnoses which are associated with management, such as pregnancy toxaemia, hypocalcaemia and urolithiasis. Fasciolosis and parasitic gastroenteritis were the main parasitic conditions diagnosed. The observed frequencies of these latter conditions - fasciolosis and parasitic gastroenteritis - were markedly raised compared to levels in 2006. This was to be expected given the wet summer in 2007, and it provides useful validation of the fluke forecasts issued in the autumn of 2006.

In both age groups, the category ‘No Diagnosis’ includes carcasses which were unsuitable for examination (invariably due to autolysis).

Figure 18: The relative frequencies of the diagnosed causes of deaths of adult sheep carcasses examined during 2007 (n = 222).
**Ovine Abortion**

Lamb foetuses are submitted to the RVLs each year to identify the cause of abortion. *Toxoplasma gondii* (33.8 per cent) was the most commonly identified agent associated with ovine abortion in 2007 - while enzootic abortion of ewes (EAE) was the second most frequently identified cause of foetal losses at 5.8 per cent (Figure 19). Neither agent can be identified by routine culture of foetal stomach contents, which is typically performed on all foetal submissions. Toxoplasmosis is diagnosed by the detection of antibodies in foetal fluids to *Toxoplasma gondii* and/or the presence of histopathological lesions of protozoal encephalitis in the brain. A diagnosis of EAE is made on the basis of a positive antibody result, histopathology, or by passage in cell culture and subsequent fluorescent antibody microscopic examination. The optimal sample for the detection of *Arcanobacterium pyogenes* (EAE) is fresh ovine placenta. The figure of 5.8 per cent is likely to be a significant underestimation of the incidence of enzootic abortion because of the failure to submit the optimal samples.

To allow use of the full range of tests, the submission of fresh placenta (including at least one cotyledon) with all foetuses - and where possible blood samples from aborted ewes - is of great importance. Likewise, the submission of samples from a number of ewes greatly enhances the probability of a definitive diagnosis being reached. A negative result from a single ewe is insufficient to rule out infectious abortion in a flock.

![Figure 19: The relative frequency of causes of abortion in sheep (n = 240).](image)

Bacterial abortions in sheep occurred with much less frequency. *Arcanobacterium pyogenes* (2.5 per cent), *Campylobacter spp.* (2.1 per cent), *Listeria spp.* (1.7 per cent) and *Salmonella dublin* (1.2 per cent) were identified in foetal stomach contents. The category 'Other Diagnoses' includes conditions such as 'toxaemia' and 'septicaemia'. All cases of abortion in ewes should be submitted to the local RVL in an attempt to identify the causative agent.

**Procedures for the submission of samples for laboratory investigation**

Compliance with the procedure for the packaging of samples being submitted to the RVL is key to protecting the health and safety of laboratory staff and postal workers. The responsibilities of the consignor are laid down in the European Agreement for Transportation of Dangerous Goods Regulations 2007 which can be viewed at:


Samples should be packaged in three layers. The primary container, which holds the specimen, should be wrapped in absorbent material and placed in a leak proof plastic container, which is then placed in the outer padded envelope or box. The words "Diagnostic Specimen" must be labelled on the outside of the package.

There are a number of suppliers of appropriate packaging material in Ireland. Contact details may be obtained from the Institute of Packaging Ireland (also known as the Irish Packaging Society).
**Parasitic diseases**

The Regional Laboratories examined a large number of samples for evidence of internal and external parasitic infections in 2007. The majority of submissions were faecal samples, coming from herds or flocks where animals were showing clinical signs of diarrhea or weight loss. In a small number of cases, faecal samples were taken post anthelmintic treatment to check on the effectiveness of the treatment.

It is important to remember that faecal egg counts are not a direct measure of the actual parasite burden of the host animal. Factors such as parasite species, stage of infection, and faecal consistency, must be taken into account when interpreting faecal egg counts. The dilution effect of faecal consistency can have an impact on counts. Paradoxically, animals with watery diarrhea due to severe parasite infestation may have relatively low faecal egg counts due to dilution. The developmental stage of the parasites can also affect counts – immature parasites generally do not produce eggs. Faecal egg counts must be interpreted with particular caution in relation to early nematodirus, type II ostertagiasis, and liver fluke infections.

**Bovine Parasites**

1,596 faecal samples (from 918 separate farms) were examined for the presence of *Fasciola hepatica* (liver fluke) eggs in 2007. 166 (10.4 per cent) were positive for fluke eggs. The farm incidence rate was 10.2 per cent. The results showed a significant increase from 2006 when only 5.3 per cent of 1,312 samples tested were found to be positive for fluke eggs.

Figure 21 illustrates the monthly results and shows that, particularly for the months April to November, the 2007 results were consistently higher than those for 2006. It is likely that the wet summer of 2007 had an impact on the number of positive results, with the conditions being more conducive to the survival of the intermediate host, the mud snail *Lymnaea truncatula*. The Department of Agriculture, Fisheries and Food issued an alert in autumn 2007 warning that liver fluke infection would be more prevalent over the 2007/2008 winter/spring period than in previous years.

Parasitic pneumonia, associated mostly with *Dictyocaulus viviparus* infection, can cause severe disease in cattle, particularly in the first grazing season. Twenty-five (4.4 per cent) of the 566 faecal samples analysed for the presence of lungworm larvae in 2007 were positive. Sixty per cent of the positive samples were received in the months of July and August.

Figure 22: The percentage of bovine faecal samples positive for strongyle eggs by month in 2006 and 2007.

Ostertagia ostertagi is the main cause of parasitic gastritis in Ireland. Type I disease typically occurs in calves during the first grazing season, with clinical signs seen 3-4 weeks after the ingestion of larvae on intensively managed farms. At this time the Ostertagia ostertagi worms are usually accompanied by other trichostrongylid worms such as *Trichostrongylus axei, Trichostrongylus colubriformis and Cooperia spp.* Type II disease is seen in older cattle, usually in the winter and spring following the first grazing season.
It is associated with the emergence of inhibited larvae from the abomasum. The eggs of *Ostertagi ostertagi* are similar to other strongyle eggs and these are grouped together for reporting purposes.

All RVLs use the McMaster flotation method to quantitatively assess faecal strongyle egg numbers. Figure 22 summarises the findings for 2007.

**Ovine Parasites**

401 ovine faecal samples were examined for the presence of liver fluke eggs. Eighteen (4.5 per cent) were found to be positive. A total of 617 ovine faecal samples were examined for the presence of strongyle eggs. Figure 23 summarises the findings.

*Nematodirus* species eggs were seen in 67 (10.8 per cent) of the samples examined. When the figures are broken down by month (Figure 24), it is apparent that there was a sharp rise in the percentage of positive samples during the summer months. It is important to point out however, that in relation to infections with *Nematodirus spp.*, the clinical signs of disease typically occur in the prepatent period, when the parasites have not yet reached the egg-producing adult stage. The recently described phenomenon of the continuation of *Nematodirus spp.* infections into the autumn and early winter is also evident from this data. This may also be attributable to the wet summer of 2007.

*Psoroptes ovis*, the non-burrowing mange mite which is the cause of sheep scab, was identified in 12 wool samples submitted from ten farms during 2007. As it is unlikely that samples are received from all positive or suspect flocks, the prevalence of sheep scab is probably significantly higher than these figures suggest.

Sheep scab is a highly contagious, chronic, insidious disease. In situations where sheep have been diagnosed with *Psoroptes ovis* mites it is important that treatment and control measures be instituted on a flock basis.
Antibiotic Sensitivity Profiles 2007

Selections of bacteria that are isolated from pathology submissions by the RVLs each year are subjected to in-vitro antimicrobial resistance testing. The results generated are of interest because resistant bacteria may cause severe infections which can be difficult for the veterinary practitioner to treat and which may also be of zoonotic significance. The panels of antibiotics used have been selected to reflect the range of antimicrobials available for the treatment of infections of farm animals.

Milk Pathogens

Bacteria isolated from milk samples are tested for resistance to a panel of antibiotics selected to include the classes available for intra-mammary and parenteral treatment of mastitis. The panel comprises compounds from 6 antibiotic classes: beta lactams (cloxacillin and penicillin), beta lactams with beta lactamase inhibitor (amoxicillin-clavulanate), cephalosporins (cephalexin), macrolides (erythromycin), aminoglycosides (neomycin, framycetin), and tetracyclines (tetracycline).

Figure 25: Antimicrobial resistance in E. coli from bovine milk samples (n = 402).

Escherichia coli

Resistance patterns for E. coli isolates from milk samples are shown in Figure 25. The results are similar to those published in the RVL surveillance report for 2006. Thirty eight per cent of strains were susceptible to amoxicillin-clavulanate, aminoglycoside, cephalosporin and tetracycline, and 14 per cent of the isolates tested were resistant to 3 or more of these 4 antibiotic classes.

Streptococcus dysgalactiae

Resistance patterns for Strep. dysgalactiae are shown in Figure 26. The results are comparable to those reported elsewhere in the scientific literature. The majority of isolates were sensitive to beta-lactams and cephalosporins. Resistance was recorded to tetracycline and to aminoglycosides.

Figure 26: Antimicrobial resistance in Streptococcus dysgalactiae from milk (n = 137). (Amc= amoxicillin clavulanate).

Enteric Pathogens

Bacteria isolated from faecal samples are tested for in-vitro antibiotic sensitivity using 6 classes of antimicrobials; beta lactams -beta lactamase inhibitors (amoxicillin-clavulanate), aminoglycosides (apramycin, framycetin, neomycin), fluoroquinolones (enrofloxacin), phenicols (florfenicol), potentiated sulphonamides (sulphamethoxazole/trimethoprim) and tetracyclines (tetracycline).
**Escherichia coli**

The great majority of *E. coli* isolates from faecal and intestinal samples are likely to be commensals and of little or no pathogenic significance. However, they can act as potential reservoirs of antimicrobial resistance genes that may be transferred to more pathogenic strains of bacteria. Significant levels of resistance to multiple antibiotics were seen in *E. coli* isolates (Figure 27). For example, 84 per cent of isolates from porcine faeces were resistant to three or more classes of antibiotics.

![Figure 27: Antimicrobial resistance in *E. coli* isolates from bovine faeces (n = 2,245). (Sxt = sulphamethoxazole-trimethoprim, Amc = amoxicillin clavulanate).](image)

**Salmonella species**

Figure 28 presents the resistance levels recorded during 2007 in *Salmonella dublin* isolates which were similar to those reported for RVL data in 2006. Almost all isolates (97.5 per cent) were susceptible to all antimicrobials on test. This is in contrast to recent US findings where significant levels of antibiotic resistance in *S. dublin* have been identified (Ray et al. 2007).

![Figure 28: Antimicrobial resistance in all *Salmonella dublin* isolates, n=143. (Amc = amoxicillin clavulanate, Sxt = sulphamethoxazole-trimethoprim)](image)
Class A Disease Surveillance 2007

The regional veterinary laboratories play an essential role in the national surveillance for exotic disease - and in supporting the state services during disease outbreaks. The strategic location of the RVLS provides a network of laboratories to cover the entire country. Animal disease surveillance is a risk control measure that reduces time to detection.

Foot and Mouth Disease

The most recent outbreak of Foot and Mouth disease (FMD) in Ireland occurred in 2001 in Co. Louth. FMD is a highly contagious disease of cloven-hoofed animals. The State Veterinary Service is vital in performing surveillance to prevent the reintroduction of the disease into the country. The outbreak in England during 2007 serves as a reminder of the need for constant vigilance. The District Veterinary Offices (DVOs) and Regional Veterinary Laboratories (RVLS) investigated four FMD suspects in four different counties in 2007. Three of these suspects were in cattle while one sheep flock was also examined. In all four cases, following joint inspections by the DVO and RVL staff, FMD was ruled out. None of the four cases required sampling or movement restrictions to be put in place.

Bluetongue

Bluetongue is an OIE listed viral disease of ruminants that has 24 known serotypes. The incubation period is between 4 and 20 days. Infection may be subclinical with no signs of illness, or may present as a clinical disease of variable severity and mortality rates of between 4 and 30 per cent. It is common to find only one or two animals affected in a herd.

References

Transmission is usually vector mediated (Culicoides midges), but may also be transmitted by the transplacental route, contaminated semen, or by iatrogenic means. The clinical signs of Bluetongue include pyrexia, anorexia, inflammation and hyperaemia of the mucosal surfaces, oedema of the tongue, lips and face, and ulceration and necrosis of the palate, tongue and nares. The full range of clinical signs is listed on the Department of Agriculture website www.agriculture.gov.ie.

The majority of Bluetongue testing in Ireland is directed at tracing imported cattle and sheep. In 2006, 373 imported cattle and 54 imported sheep were tested by Bluetongue ELISA and proved negative. This number rose to 1,239 imported cattle in 2007 - all of which proved negative. In addition, a random national survey is conducted annually to prove freedom from the disease. In 2007, 1,375 cattle from 275 herds were tested for Bluetongue antibody and found to be negative (Figure 30). The Brucellosis Laboratory is currently collecting samples for the 2008 random national survey.

A total of 1,245 samples from birds was tested in 2007 for Avian Influenza by real-time PCR (Table 7). Only one specimen was positive - a non-pathogenic H6N2 virus isolate from chickens in Cork. Submissions were initially examined in the RVLS by laboratory staff and samples were selected for analysis by Virology Division in the Central Veterinary Research Laboratory (CVRL) at Backweston. Ireland’s continuing freedom from pathogenic avian influenza is dependant on the constant vigilance of the public and flockowners for signs of avian influenza. More information is available on www.agriculture.gov.ie.

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</tr>
<tr>
<td>Poultry</td>
<td>543</td>
<td>1 (H6N2)</td>
</tr>
<tr>
<td>Captive</td>
<td>143</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>1245</td>
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</tr>
</tbody>
</table>

Table 7: Testing conducted for Avian Influenza in 2007.

Avian Influenza

The spread in recent years of the highly pathogenic avian influenza from Asia to the west has provoked great concern in the EU. This disease can have devastating consequences for the health of birds - and can sometimes pose a threat to human health. In response to the risk of introduction of Avian Influenza into Ireland, the Department of Agriculture, Fisheries and Food has implemented preventive and control measures to ensure early detection of infected birds and to help contain the disease in the event of an outbreak. Concerns regarding deaths or illness in wild or domestic birds can be reported by calling the Department’s hotline on 1890 252 283.

Figure 31: Hyperaemia and oedema of the lips, nose and mouth in a ewe seven days post infection with Bluetongue Serotype 8. Reproduced from Darpel, K.E. et al., Veterinary Record (2007) 161, 253-261 with the kind permission of the Veterinary Record.

A Selection of RVL Farm Investigations in 2007

The Regional Veterinary Laboratory service provides a comprehensive laboratory-based diagnostic pathology service to herd owners through their veterinary practitioners. Laboratory pathologists are also available for consultation and for on-farm investigation of animal disease and production problems. Summaries of selected farm investigations carried out by RVL staff during 2007 are presented below.

Fertility Problems

Kilkenny RVL investigated a farm with a history of undiagnosed stillbirths for four years. In 2007, 16 of 32 heifers delivered full-term dead calves, and another two aborted at eight months. Many of the heifers were sick during their first lactation, some with respiratory disease and some with left displacement of the abomasum. Five had to be dried off, and two died. The herd owner also reported severe enteritis in young calves and pneumonia in purchased weanlings. Eight foetuses were submitted for laboratory examination and blood samples from the dams and their cohorts were examined serologically and biochemically. Maternal serology revealed a low incidence of neosporosis (3/34) and a high prevalence of BVD antibodies (52/56). However, no pattern was discovered to explain the foetal deaths. A blood screen of dams and cohorts revealed marginal calcium (5/32) and magnesium levels (7/32). Subclinical hypocalcaemia has been associated with slow calving, and also with postparturient diseases. Laboratory analysis identified low immunoglobulin levels (ZST), as well as Rotavirus and Cryptosporidium infection, in the young calves.
Nutritional alterations to rectify hypocalcaemia, as well as general advice on disease management in the calves, resulted in a marked improvement in herd health the following season.

**A high prevalence of milk fever – two investigations**

A 70-cow dairy herd was referred to Sligo RVL to investigate an increased prevalence of milk fever. At the time of the investigation, there were five cases of milk fever in 30 calved cows, two of which died. The herd was using a DCAB (Dietary cation-anion balance) strategy to prevent milk fever. This concept focuses attention on the level of potassium that is contained in the feed of dairy cows. It is widely accepted that the homeostatic mechanisms that result in milk fever prevention work best when DCAB is negative. The pH of the urine of a number of dry cows, close to full term, was examined and found to be more alkaline than it should have been. High potassium levels in the silage were suspected of causing the alkalosis. Magnesium levels were low to marginal. High potassium in ruminal fluid can reduce magnesium uptake from the diet. The problem was addressed by supplementing the dry cow diet with 30g of calcined magnesite.

Athlone RVL investigated a 130-cow dairy herd with a history of milk fever pre-calving – which responded poorly to calcium treatment. Animals that did respond were reported to subsequently suffer setbacks in their condition. A large number of cows (12 per cent) had failed to hold to service and retained foetal membranes and left-displaced abomasum (LDA) were ongoing problems. Blood samples were taken for mineral analysis, and to screen for infectious causes of foetal loss. Of particular note was the level of phosphorus, which was low in 50 per cent of those analysed – despite the use of Vitamin D3 injections to promote the intestinal uptake of phosphorus. No evidence of associated anaemia was evident from the haematology results. The milk fever problem appeared to be due to both calcium and phosphorus imbalances – with the latter giving rise to poor response to treatment. Supplementation of phosphorus was undertaken immediately. Advice was also given regarding conducting a soil analysis to determine if fertiliser usage needed to be reviewed. A reassessment of the diet used, and the use of precalver minerals, were advised to prevent a recurrence.

**Neonatal mortality in a suckler herd.**

Sligo RVL was asked to investigate neonatal mortality in a suckler herd. At the time of the visit, seven of the 32 cows calved had lost their calves. According to the herdowner, calves became ill and died following a short clinical course. One calf was submitted to Sligo RVL for post mortem examination prior the visit. This calf had a severe extensive fibrinous peritonitis. Its ZST reading was 2 units, indicating it had not received colostrum. According to the history, cows had been administered an enteric vaccine during the dry period. The farm visit identified some problems with housing – including absence of a dedicated area for calves. Advice was given regarding the importance of ensuring calves receive adequate colostrum immediately after birth, and some changes in calf housing use and layout were recommended.

**Lameness in store cattle**

In the autumn, Dublin RVL investigated a chronic lameness problem in a group of store cattle. The problem appeared approximately 3 weeks after housing in a slatted shed in early September. At the time of the visit, 10 cattle had been affected, and while most had recovered, two remained chronically lame. Examination of affected animals revealed clinical symptoms consistent with interdigital necrobacillosis (‘Foul in the Foot’). Powerhosing of the housing and disinfectant foot bathing were recommended, while the importance of adequate antibiotic therapy in clinical cases was also stressed.

**An outbreak of coccidiosis in calves**

An investigation was carried out by Athlone RVL, in the spring of 2007, into a problem of diarrhoea in calves from one month of age. According to the herdowner, the problem was first noticed in 2006, when diarrhoea continued until July. Some of the affected calves from that year were subsequently stunted. Coccidiosis was suspected. Although faecal samples were submitted to the laboratory at the time, they were negative for coccidial oocysts. On-farm, calves at about 6 weeks of age were placed in an old calfhouse with a low roof and walls which were difficult to clean. This was where the diarrhoea normally started. During the field investigation, samples were collected from untreated calves with diarrhoea. These were positive for coccidial oocysts. Advice was given concerning early coccidial prophylaxis, and avoiding the use of contaminated sheds. Feedback from the farm confirmed that this course of action has been successful. This case underlines the importance of sampling the correct animals at the right time to yield positive results. It also highlights the role of unsuitable housing in the incidence of disease – and the impact which diseases such as coccidiosis can have on growth performance of calves.
A Selection of Abstracts from Published Papers 2007

Stunting in cattle on three farms associated with calfhood disease

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(Reproduced with their kind permission)

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Three herds, located in widely separate rural locations in Ireland and having a high incidence of stunting, were investigated by the Regional Veterinary Laboratory Service (RVLS). Some animals required approximately a year longer to reach slaughter weight or first lactation while others never reached mature size. Stunted animals on all farms were neither depressed nor inappetant and the herd owners did not consider them clinically ill. On each farm, stunted animals were clinically examined and samples were taken for laboratory analysis. All farms had been recognised for some time as being geochemically susceptible to mineral deficiencies and appropriate mineral supplementation was routinely practised. In the three herds described, growth retardation in calves commenced and continued during prolonged and repeated septicaemic, enteric and respiratory disease episodes while housed. Lung damage in many animals persisted long after apparent clinical recovery. Compensatory gain did not occur after apparent clinical recovery resulting in a lower growth trajectory and thus resulting in stunting persisting into later life. The authors conclude that current development on farms of large calf rearing units without proper isolation facilities makes it likely that this syndrome will become a more frequent occurrence.

Severe facial oedema associated with orf in an Irish sheep flock

(Reproduced with their kind permission)

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The authors described an atypical presentation of contagious pustular dermatitis (orf) in a sheep flock in the west of Ireland, in which extensive facial oedema was a major presenting feature. The affected flock consisted of 55 Texel cross lambs, aged three to four months. Orf had occurred on this farm in previous years but no vaccination strategy was in place for the disease. Lambs presented with pustules and encrusted nodules on the lips and nares, which extended in some cases over the muzzle. Twenty-four of the affected lambs also exhibited marked ‘pitting’ oedema in the soft tissues of the face and the submandibular lymph nodes were palpably enlarged. A presumptive diagnosis of orf was confirmed by detection of parapox virus particles on electron microscopic examination of scab material. The lambs were treated to prevent secondary bacterial infection. When the flock was revisited 10 days later, all of the sheep that had been affected were recovering from orf and the facial oedema had completely dissipated. The superficial resemblance of the affected lambs in this case to recently published photographs of experimental Bluetongue cases (see Figure 31, page 18) serves as a timely reminder that orf has a wide spectrum of clinical presentations and is an important differential diagnosis for suspected cases of Bluetongue.

Figure 32: Facial oedema in a Texel ewe associated with orf.
(Photo: Micheál Casey).