

# Regional Veterinary Laboratories - Surveillance Report 2009



Department of  
**Agriculture,  
Fisheries and Food**

An Roinn  
**Talmhaíochta,  
Iascaigh agus Bia**

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## Contents

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<b>Introduction</b>	2	<b>Clinical Chemistry</b>	30
<b>Overview of RVL Submission Rates, Animal Demographics and Weather</b>	3	Copper and Selenium Analyses in the Regional Veterinary Laboratories in 2009	30
Submission Rates to the Regional Veterinary Laboratories in 2009	3	Haematology Testing in the Regional Veterinary Laboratories	31
Animal Demographics in 2009	3	Tick-borne Fever in a dairy herd	31
Weather in 2009	4	Proficiency Testing in the Regional Veterinary Laboratory Service	31
<b>Diseases of Cattle</b>	5	Procedures for the submission of samples for laboratory investigation	32
Neonatal Calves (birth to one month)	5	<b>Class A Disease Surveillance 2009</b>	32
Congenital Joint Laxity and Dwarfism in Calves (CJLD)	6	Foot-and-Mouth Disease	32
Calves (one to three months)	6	EuFMD Training in Erzurum, Turkey	33
Weanlings (three months to one year)	7	Bluetongue	33
Adult Cattle	8	Avian Influenza	33
Clostridial Diseases in Cattle	9	Bovine Spongiform Encephalopathy (BSE)	34
Poisonings in Cattle	9	Scrapie	34
Bovine Neonatal Enteritis	10	<b>A Selection of RVL Farm Investigations in 2009</b>	35
Bovine Neonatal Pancytopenia – A Novel Disease	12	Mortality in a sheep flock due to Paramphistomosis	35
Bovine Abortion	13	Immunosuppression in a suckler herd	36
Bovine Mastitis	15	Actinobacillosis in a herd of bullocks	36
Bovine Respiratory Disease	16	Psoroptic mange in a Belgian Blue bull	36
Johne's Disease 2009	18	An investigation into suspected triclabendazole-resistance in a sheep flock with chronic liver fluke disease ( <i>Fasciola hepatica</i> )	37
Biosecurity	19	An infertility problem in a dairy herd	38
<b>Diseases of Sheep</b>	20	<b>A Selection of Abstracts from Published Papers 2009</b>	38
Mortality in lambs (birth to six months)	20	Nephrotic syndrome due to glomerulopathy in an Irish dairy cow	38
Mortality in adult sheep (over six months)	20	The ovine placenta and placentitis - A review	39
Ovine Abortion	21	Avian influenza viruses detected by surveillance of waterfowl in Ireland during 2003-2007	39
<b>Diseases of Pigs</b>	22	Commensal rodents at farm yards in Co. Cork	40
Postweaning Multisystemic Wasting Syndrome	22	Demographics of cattle positive for <i>Mycobacterium avium</i> subspecies <i>paratuberculosis</i> by faecal culture, from submissions to the Cork Regional Veterinary Laboratory	40
Accidental Selenium Intoxication in a Pig Herd	23		
Swine Influenza in a Pig Herd	23		
<b>Parasitic Diseases</b>	24		
Liver Fluke and Rumen Fluke Infections	24		
Gastro-intestinal Parasitic Infections	25		
Lungworm Infections	26		
The <i>Trichinella</i> spp. Survey 2009	26		
<b>Antimicrobial Resistance Profiles 2009</b>	27		
Milk Pathogens	27		
Enteric Pathogens	28		

## Introduction

This is the fifth Annual Report on the animal disease surveillance activities of the Department of Agriculture, Fisheries and Food (DAFF) Regional Veterinary Laboratories. The Regional Veterinary Laboratories (RVLs) comprise six diagnostic pathology laboratories in Athlone, Cork, Dublin, Kilkenny, Limerick and Sligo and are an integral part of the Veterinary Laboratory Service (VLS). They are strategically located around the country to provide optimal coverage of the national farm animal populations. This report provides an overview of their findings during 2009. The material derives from analysis of the results of examinations on carcasses and clinical pathology submissions, as well as the results of on-farm investigations by laboratory staff.

The RVLs occupy a central role in the national animal disease surveillance programme. A vigilant national surveillance network is vital in identifying changes in the trends and distribution of animal disease - thereby facilitating disease control. The unique collaborative links which have been fostered between the RVLs and the farming community, through private veterinary practices, promote the existence of an effective and sensitive passive surveillance network. Clinical pathology submissions from live animals (bloods, swabs, faeces, etc.), or carcasses of dead animals, are received in the RVLs from private veterinary practitioners (PVPs) on a daily basis. The investigations conducted in the RVLs by veterinary pathologists and laboratory analysts provide a unique source of data on disease occurrence in the national farm animal population. This helps inform decisions on animal health strategy - both at national and at farm level.

The favourable health status of the Irish national herd is something of which we are justifiably proud. However, for an industry which relies heavily on the export sales of farm produce, the risk of the introduction of new or exotic diseases into the country is an ever present threat to the agricultural industry as well as to the economy at large. The RVLs have an important role in the rapid recognition of new or exotic diseases within Ireland, as well as in monitoring the occurrence of endemic diseases - in particular those which have significant impacts on farm productivity and animal welfare. A selection of findings from the results of these surveillance activities are included in this report.

The RVLs also fulfil an important advisory role to the farming community through their private veterinary practitioners. Veterinary pathologists are available in the RVLs for consultation on unusual disease presentations, epidemiological advice, and also to carry out on-farm investigations. Summaries of a selection of these investigations are outlined on page 35 of this report.

The VLS Laboratory Information Management System (LIMS) is an integrated data management system which has been used in the RVLs since 2002. It links the regional and central laboratories in real time and records all submission information from sample receipt, through testing, to authorisation and final reporting. This central database allows details relevant to individual animal disease investigations to be recorded, and facilitates easy access to data on disease trends and distribution across the RVL network.

It is envisaged that this report will be of value to those involved in the protection and promotion of animal health and welfare in Ireland. It provides summary highlights of a key component of the Department's program of surveillance for animal disease, and is an important source of data on the causes of illness and deaths in Irish livestock.

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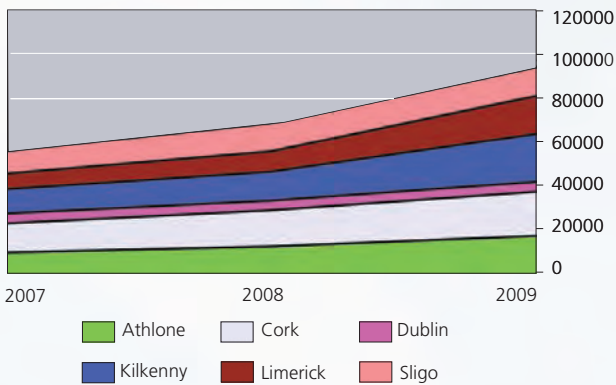
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## Overview of RVL Submission Rates, Animal Demographics and Weather

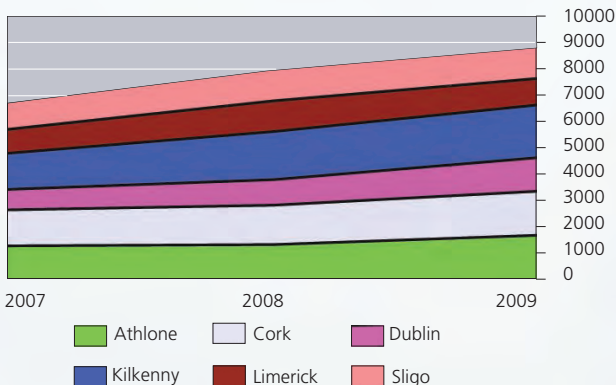
### Submission Rates to the Regional Veterinary Laboratories in 2009

In 2009, the Regional Veterinary Laboratories (RVLs) analysed a total of 97,085 diagnostic samples and examined a further 8,824 carcasses. The number of diagnostic samples submitted to the RVLs has consistently increased over the last number of years. The 2009 figure represents an increase in diagnostic submissions of 66.1 per cent in the last three years alone ( Figure 1).



**Figure 1: Trends in the submission of clinical pathology samples to the Regional Veterinary Laboratories over the three years 2007 to 2009 <sup>1</sup>.**

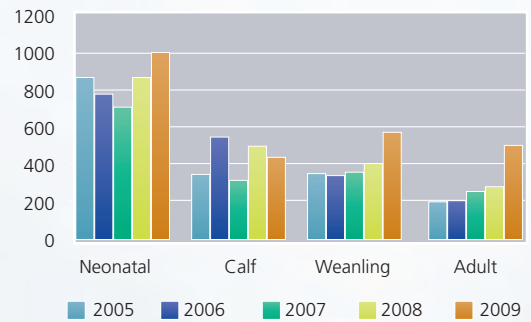
The numbers of carcass submissions to the RVLs have also increased in the same period, albeit at a lesser rate, with an increase of 32.1 per cent recorded in the last three years (Figure 2).



**Figure 2: Trends in the submission of carcasses for post mortem examination to the Regional Veterinary Laboratories over the three years 2007 to 2009.**

These figures reflect an increasing awareness of the value of diagnostic pathology in the diagnosis, prevention and treatment of animal disease. It possibly also reflects the change in focus within veterinary practice from the treatment of individual animals to herd health programmes, in which laboratory testing to determine the herd health status plays a central role.

<sup>1</sup> Dublin RVL numbers exclude parasitological and clinical chemistry submissions.

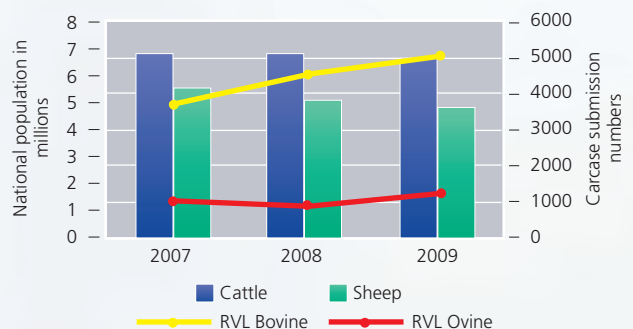


**Figure 3: The number of bovine carcass submissions, by age category, submitted to the six RVLs in the years 2005 to 2009.**

Figure 3 illustrates a breakdown of bovine carcass submissions to the RVLs by age category over the last five years. The numbers of carcass submissions have increased in each age category with the exception of the calf category (one to three months of age). Neonatal calves (birth to one month of age) are consistently the most common bovine carcass submission to the RVLs which underlines the significant losses in this age category on Irish farms - many of which can be prevented by appropriate management (see 'Bovine Neonatal Enteritis', page 10). There was a significant increase in 2009 in the numbers in the adult and weanling (three months to twelve months of age) categories in particular. Further details on the findings in bovine carcass submissions are presented on page 5.

### Animal Demographics in 2009

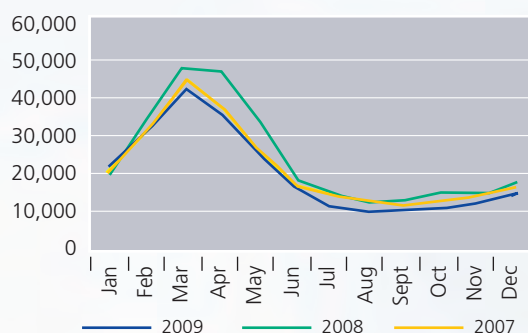
Figure 4 shows the national total populations of cattle and sheep as measured in June of each year for the years 2007, 2008 and 2009 compared with the carcass submission rates for cattle and sheep carcasses to the Regional Veterinary Laboratories in the same period.



**Figure 4: Total cattle and sheep numbers, recorded in millions, in Ireland as measured in June each year and the bovine and ovine carcass submission rates for 2007, 2008 and 2009 (National data sourced from the Central Statistics Office <http://www.cso.ie>).**

The national cattle population remained quite constant during this period, while sheep numbers fell by 13.4 per cent from 5.52 to 4.78 millions. Against this backdrop, bovine carcass submission numbers to the RVLs increased by 37.6 per cent, and ovine carcass submission numbers increased by 15.2 per cent over the same period.

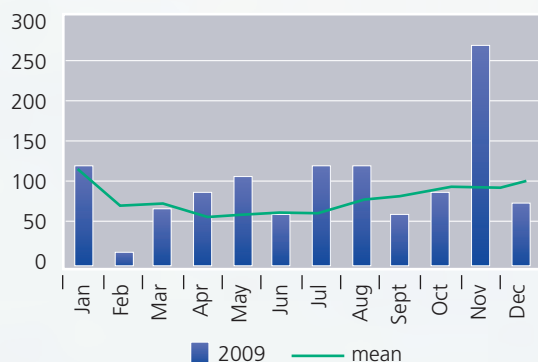
Total numbers of on-farm cattle deaths by month as recorded in the DAFF Animal Identification and Movement (AIM) Bovine Statistics Reports for the three years 2007, 2008 and 2009 are shown in Figure 5. They follow an expected seasonal pattern with the period of highest mortality coinciding with the peak calving season. Mortality figures for 2009 were consistently lower than those for 2008, and mirror closely those recorded in 2007.



**Figure 5: The total numbers of cattle deaths (cattle and aborted foetuses) recorded on the Animal Identification and Movement (AIM) system by month for the years 2007, 2008 and 2009 (Source: DAFF AIM Bovine Statistics Reports).**

## Weather in 2009

The most notable features of the Irish weather in 2009 were the heavy Summer and November rainfalls (Figure 6), and the low daily temperatures experienced in December (Figure 8). Of these, the heavy rainfall appeared to have the greatest impact on animal disease, with a large increase recorded in the diagnoses of *Fasciola hepatica* related deaths and the emergence of larval paramphistomosis as a clinical entity in juvenile cattle and sheep in October and November (see 'Diseases of Cattle' and 'Diseases of Sheep' sections). In addition, there was increased detection of *Fasciola hepatica* and *Paramphistomum* spp. eggs on faecal parasitological examinations from live animals (see 'Parasitic Diseases' section).



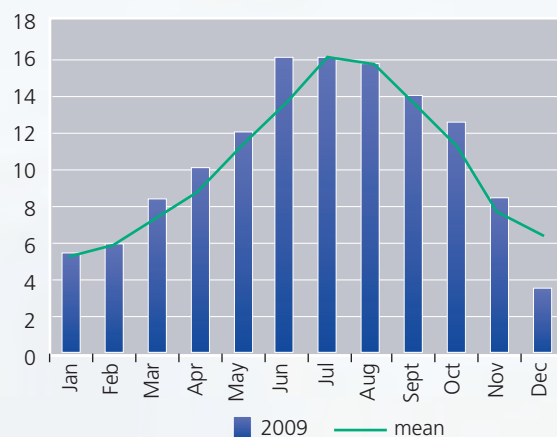
**Figure 6: The average monthly rainfall measured at Shannon Airport during 2009 compared to the mean monthly rainfall for the years 1961-1990 (Data courtesy of Met Eireann, <http://www.met.ie>).**

The risk of liver fluke and rumen fluke is closely linked to summer rainfall amounts. Wet and warm summers promote *Fasciola* development and survival, as well as providing the optimal conditions for survival of the intermediate host, the snail.



**Figure 7: Saturation of the soil was a feature of the early summer in 2009. Poached ground provides suitable temporary habitats for the snail *Galba truncatula* (formerly known as *Lymnaea truncatula*), the intermediate host of liver fluke.**

Heavy rainfall experienced in April and May resulted in delayed turnout of animals to pasture. In some areas of the country, re-housing of animals was necessary in July due to saturation of the soil (Figure 7). Both of these events may also have contributed to higher levels of diseases which tend to be associated with housing such as calf enteritis (see Figure 27) and bovine mastitis (see Figure 38).



**Figure 8: The average monthly temperature measured in Shannon Airport during 2009 compared to the mean monthly temperature for the years 1961-1990 (Data courtesy of Met Eireann, <http://www.met.ie>).**

Mean monthly temperatures for 2009 are shown in Figure 8. December was exceptionally cold. The cold temperatures experienced in December would be expected to have had an adverse effect on fluke survival over the winter as both the multiplication and shedding of cercariae and multiplication of snails is inhibited; however initial indications from early 2010 are that the risk period for liver fluke infection had a later onset, rather than the risk being reduced. The reason for this is not immediately apparent.

## Diseases of Cattle

In the face of significant increases in carcase submissions to the RVLs, the causes of deaths of different categories of cattle remain remarkably consistent from year to year.

The most striking difference in 2009 compared to previous years was the number of cattle deaths attributed to fasciolosis (liver fluke). Five per cent of weanling deaths were attributed to this condition in 2009 compared to 0.26 per cent in 2008. Among adult cattle, fasciolosis accounted for 9 per cent of adult deaths in 2009 compared to 0.37 per cent in 2008.

### Neonatal Calves (birth to one month)

As in previous years, enteric infections were the most common causes of deaths in neonatal calves, followed by septicaemia or bacteraemia, and respiratory infections (Figure 9).

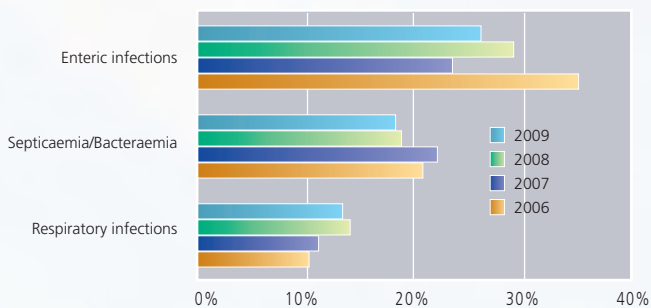


Figure 9: A comparison of the most frequently diagnosed conditions in calves from birth to one month of age for the years 2006 to 2009.

*E. coli* (43 per cent) and *Salmonella* Dublin (26 per cent) were the pathogens most frequently isolated in cases of septicaemia/bacteraemia (Figure 10).

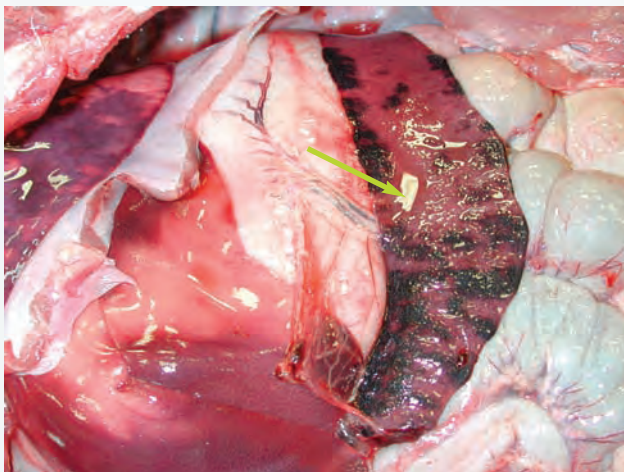


Figure 10: Diffuse haemorrhages in the spleen (arrow) of a 10 day old calf associated with *Salmonella* Dublin septicaemia (Photo: Ann Sharpe).

The category of 'hereditary and developmental abnormalities' includes a condition referred to in the literature as congenital joint laxity and dwarfism. This condition was seen more frequently in the laboratories in 2009 than in previous years (see Figure 13).

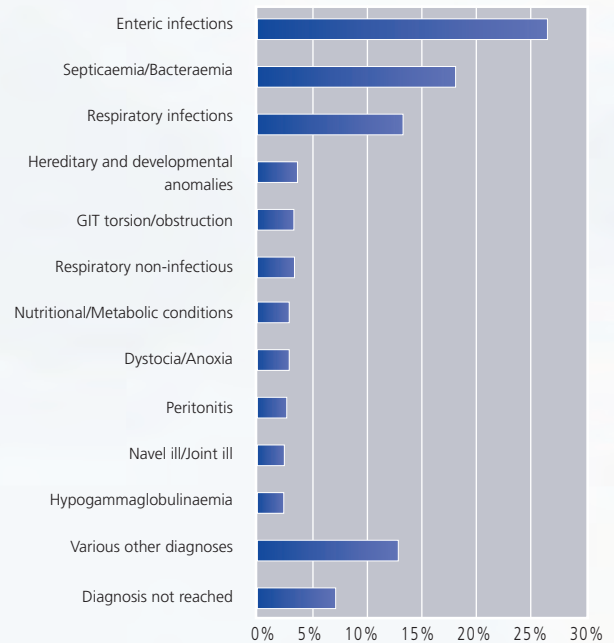


Figure 11: The most commonly diagnosed causes of mortality in calves from birth to one month of age (n=995).

Aspiration pneumonia accounts for the majority of the 'respiratory non-infectious' category of diagnoses (see Figure 11).

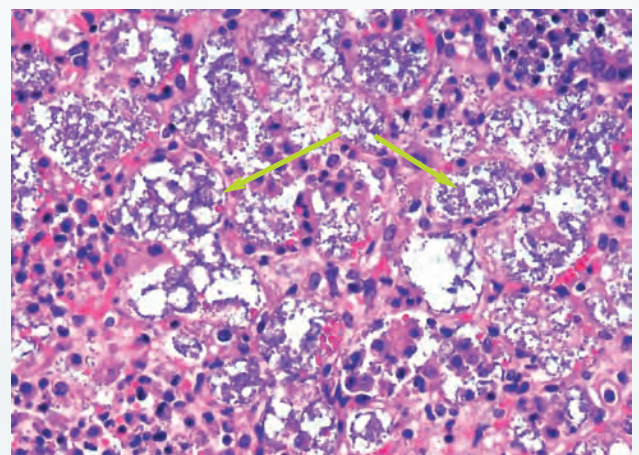


Figure 12: Photomicrograph of the lung of a calf diagnosed with aspiration pneumonia showing blue staining material (milk) containing fat globules in the alveoli (arrows) (Photo: John Fagan).

Twenty eight diagnoses of aspiration pneumonia (Figure 12) were recorded in 2009, of which twenty five were under 10 days of age. This may reflect incorrect stomach tube use. It is important that operators learn to use a stomach tube correctly before using it to administer fluids.

Inanition accounted for a number of deaths of young calves. These cases are included in Figure 11 in the 'Nutritional/Metabolic conditions' category. This is usually associated with over-zealous withholding of milk from calves during and after episodes of diarrhoea.

### Congenital Joint Laxity and Dwarfism in Calves (CJLD)

In recent years the Regional Veterinary Laboratories (RVLs) have examined calves from numerous herds which have experienced outbreaks of dwarfism - and have received reports from private veterinary practitioners regarding many other cases. Congenital Joint Laxity and Dwarfism in Calves (CJLD) encompasses a range of conditions which include congenital joint laxity, limb deformities and disproportionate shortening of the long bones, or dwarfism (Figure 13). While the condition has been recognised for many years, the exact cause has been the focus of much debate. Indeed some researchers have taken issue with the name of the condition contending that the apparent joint laxity is, in fact, due to hyperflexion caused by the disparity between long (or normal) tendons and shorter bones rather than actual joint laxity.

The condition is most prevalent in beef herds, where pit silage makes up the bulk of the winter ration, and particularly in those herds which are exposed to an extended period of feeding on winter rations due to geographical position or climatic conditions. It has also been noted that affected calves are most commonly born late in the Spring calving season or early in the Autumn calving season. These observations have pointed towards an association with the feeding of pit silage in the first half of pregnancy.



Figure 13: A dwarf newborn calf with hind limb deformity and laxity (Photo: Dónal Toolan).

In the first half of pregnancy, the growth plates in the long bones are affected as they are formed. This change is permanent after the sixth month of pregnancy has been reached. The severity of the condition in calves can vary considerably - from calves with mildly 'sickled' hocks, to severe rotation and deformity of the limbs,

necessitating euthanasia in many cases. Manganese is a mineral which is necessary for endochondral bone growth, and manganese deficiency has been suggested as a possible cause of the condition. The bioavailability of manganese in pit silage appears to be less than in other forages, which may explain how the condition arises. Alternatively, it has been suggested that a grass-silage associated teratogen may be responsible (Mee, 1995<sup>2</sup>). Preventing recurrence of the condition in the subsequent calving season is best achieved by reducing the silage content of the winter diet of cows to provide no more than 75 per cent of the total dry matter supply. This can be done by substituting with hay, straw or similar feed (Gunn *et al.*, 2000<sup>3</sup>). Following this regime has resulted in significant reductions in the incidence of the condition in affected herds.

### Calves (one to three months)

Respiratory infections, followed by enteric infections and septicaemia or bacteraemia, were the most frequently diagnosed causes of death in calves of one to three months of age examined in the RVLs in 2009 (Figure 14).

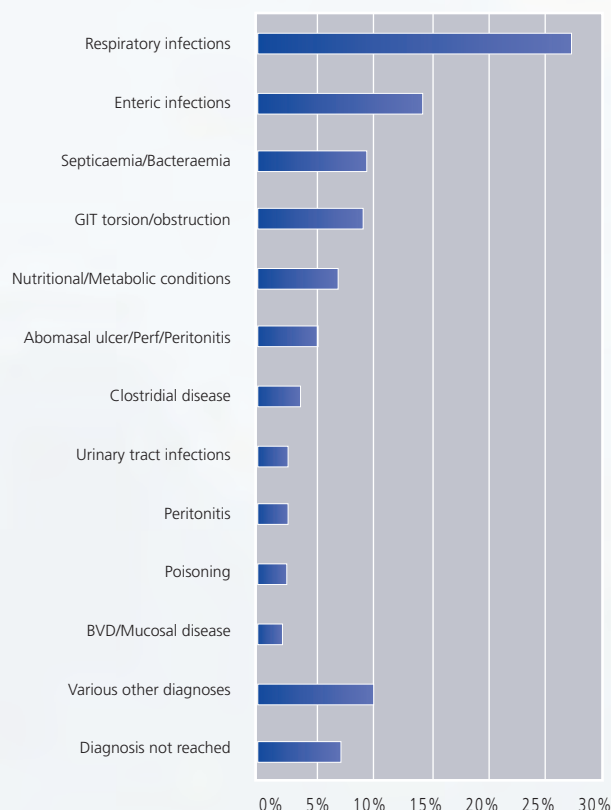


Figure 14: The most commonly diagnosed causes of mortality in calves between one and three months of age (n=442).

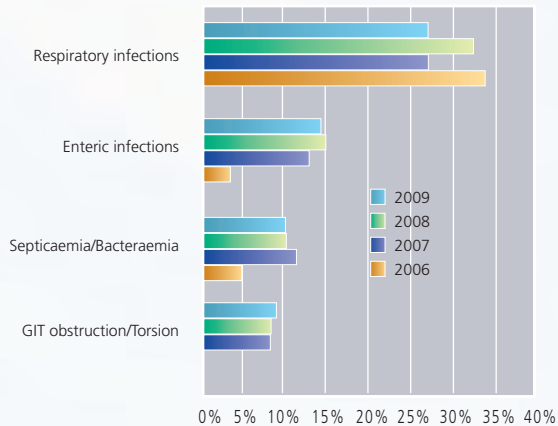
<sup>2</sup> Mee, J (1995). Nonhereditary disproportionate dwarfism in a beef herd: Clinical and pathological features. *Irish Veterinary Journal* 48: 93-105.

<sup>3</sup> Gunn, G, Caldwell G, Lowman, B (2000). Congenital joint laxity and dwarfism in calves. *Veterinary Record* 147: 199-200.



**Figure 15: Terminal gangrene in the limb of a two month old calf following *Salmonella* Dublin bacteraemia (Photo: John Fagan).**

Similar to the findings in neonatal calves, *E. coli* (12 per cent) and *Salmonella* Dublin (12 per cent) were the pathogens most frequently isolated in cases of septicaemia or bacteraemia (Figure 15). The category 'GIT obstruction/torsion' includes gastric, intestinal and mesenteric torsions as well as intussusceptions. Inanition and ruminal acidosis feature prominently in the conditions recorded as 'Nutritional/Metabolic conditions'.

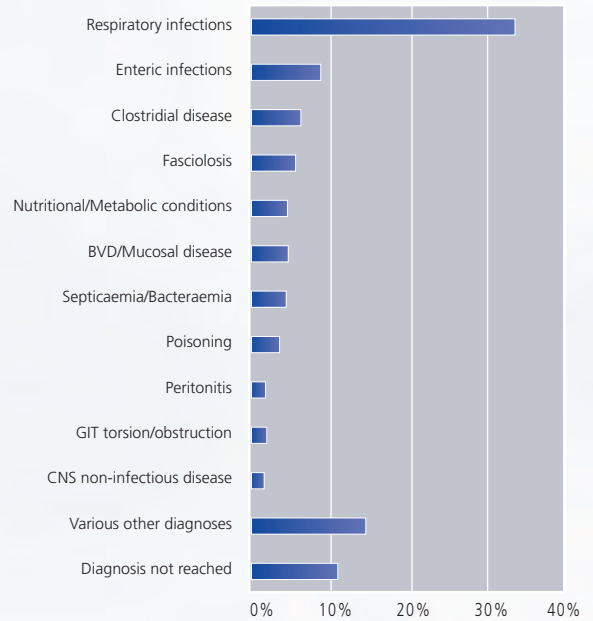


**Figure 16: A comparison of the four most frequently diagnosed conditions in calves between one and three months of age for the years 2006 to 2009.**

The relative frequency of the four most common diagnoses in calves in this age group has remained broadly similar over the last number of years (Figure 16).

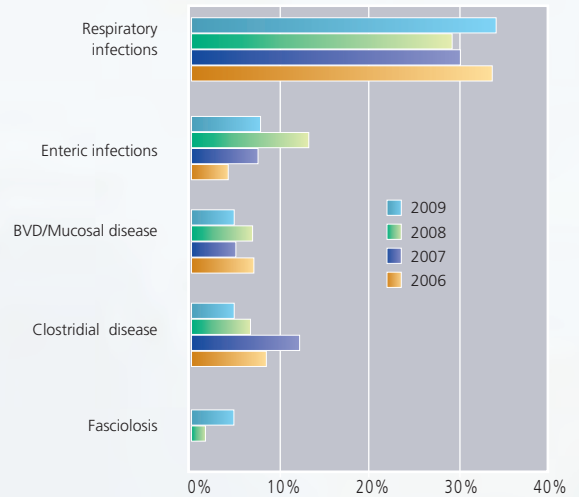
### Weanlings (three months to one year)

Fasciolosis was a significant cause of mortality among weanlings in 2009. This was most probably attributable to the adverse weather conditions and heavy rainfall through much of the summer of 2009 (see Figure 6). It was the fourth most common cause of mortality in this age group. The three most frequent diagnoses were respiratory infections (34 per cent), enteric infections (8 per cent), and clostridial disease (6 per cent) (Figure 17).



**Figure 17: The most commonly diagnosed causes of mortality in weanlings (n=575).**

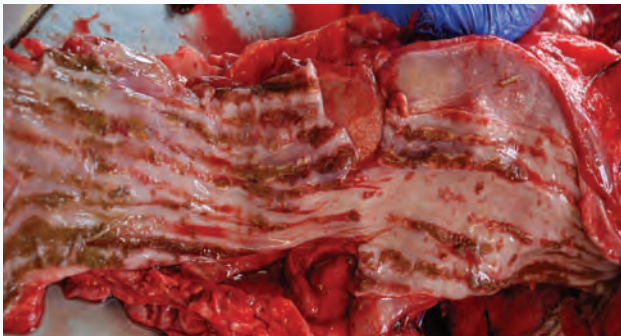
*Salmonella* Dublin (29 per cent) was the pathogen most frequently isolated in cases of septicaemia or bacteraemia.



**Figure 18: A comparison of the most frequently diagnosed conditions in calves between three months and one year of age for the years 2006 to 2009.**

The comparison of data from the years 2006 to 2009 (Figure 18) illustrates the change in fasciolosis diagnoses. No diagnoses of fasciolosis were recorded in weanlings in 2006 and 2007.



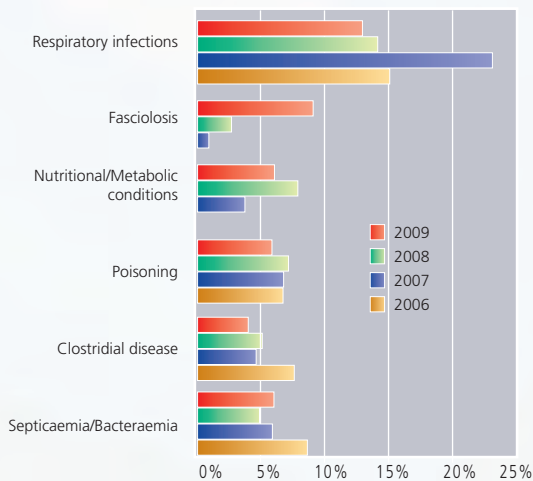


**Figure 19: Multifocal linear oesophageal ulceration caused by BVD infection in a weanling (Photo: Cosme Sánchez-Miguel).**

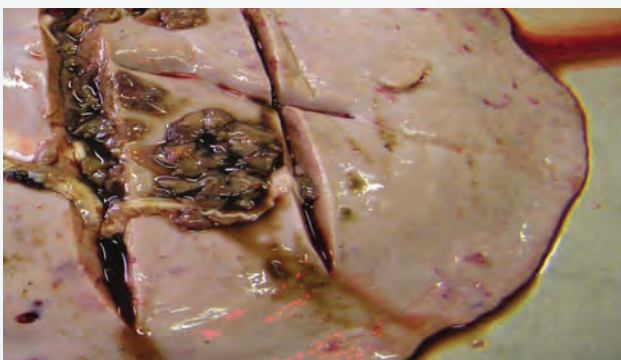
The frequency of diagnosis of BVD/mucosal disease (Figure 19) as a cause of death in weanlings in 2009 was almost half of that recorded during 2008 (Figure 18). This result is encouraging and possibly reflects an increasing awareness of the importance of identifying and removing animals persistently infected with the BVD virus from herds at an early stage<sup>4</sup>.

## Adult Cattle

The numbers of adult cattle carcasses submitted to the RVLs for *post mortem* examination almost doubled in 2009 (500 carcasses) compared to 2008 (267 carcasses).



**Figure 20: A comparison of the most frequently diagnosed conditions in adult bovines in the years 2006 to 2009.**



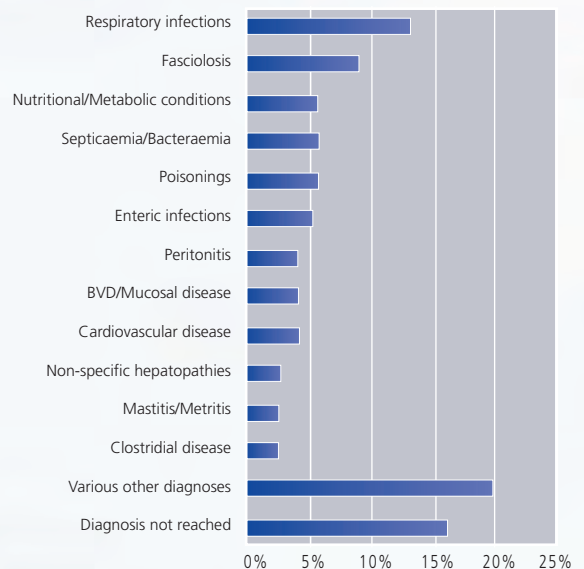
**Figure 21: Severe chronic liver fluke (*Fasciola hepatica*) infestation in the liver of an emaciated 2 year old bullock (Photo: Dónal Toolan).**

<sup>4</sup> It is important that persistently infected animals are not offered for sale or retained for breeding purposes.

When data on adult bovine mortality for the last four years is compared, the relative frequencies of the most common causes of mortality are quite similar (Figure 20) - with the exception of fasciolosis (Figure 21) which accounted for very few deaths among adult bovines until 2009.

Deaths related to Clostridial infections, at 4.4 per cent of the total, were lower in 2009 than in previous years. This marks an overall downward trend from the level of almost 8 per cent of deaths which were attributed to Clostridial disease in 2006.

Respiratory infections were the most frequently diagnosed cause of deaths in adult bovines in 2009 (Figure 22). The category 'Nutritional/Metabolic conditions', the third most frequently diagnosed cause of deaths, include conditions such as ruminal acidosis, hypomagnesaemic tetany and fatty liver disease.



**Figure 22: The most commonly diagnosed causes of mortality in adult bovines (n=500).**

The category entitled 'Cardiovascular disease' includes conditions such as endocarditis, pericarditis, myocarditis (Figure 23), and posterior vena cava thrombosis.



**Figure 23: Focally extensive myocarditis, from which *Arcanobacterium pyogenes* was isolated, in a 14 month old heifer (Photo: Ger Murray).**

Among adult cattle presented for *post mortem* examination, a wider range of disease entities were recognised than for younger cattle. Those conditions which accounted for five *per cent* or less of adult bovine deaths are included in the category entitled 'Various other diagnoses'. Among the diagnoses included in this category are gastro-intestinal torsion or obstruction (2 *per cent*), haemorrhage (2 *per cent*), babesiosis (1.2 *per cent*), CCN (0.8 *per cent*) and malignant catarrhal fever (0.6 *per cent*).

## Clostridial Diseases in Cattle

Clostridial disease accounted for three *per cent* of diagnoses in all age groups in 2009 (Table 1). Blackleg (Figure 24) was the most frequent diagnosis - accounting for 40 of the cases. Twenty seven of these were in weanlings.



**Figure 24:** A section of the *semimembranosus* muscle from a weanling showing the classical Blackleg lesion of acute locally extensive haemorrhagic myositis (Photo: Jim O'Donovan).

Seven diagnoses of Black Disease (Infectious Necrotic Hepatitis), of which six were in adults, and one diagnosis of Bacillary haemoglobinuria, were recorded in 2009. Neither Black Disease, nor Bacillary haemoglobinuria were diagnosed in 2008. *Clostridium chauvoei* (33), *Clostridium sordellii* (16), *Clostridium septicum* (5) and *Clostridium novyi* (4) were the most frequently detected clostridial pathogens.

Diagnosis	Neonatal	Calf	Weanling	Adult	Totals
Blackleg	2	9	27	2	40
Enterotoxaemia	1	2	2	6	11
Black Disease	-	-	2	6	8
Malignant Oedema	-	1	1	3	5
Septicaemia	-	1	3	1	5
Abomasitis-emphysematous	1	2	-	1	4
Peritonitis	-	-	-	2	2
Bacillary haemoglobinuria	-	-	-	1	1
Total	4	15	35	22	76

**Table 1:** The frequency of detection of clostridial disease in cattle in 2009, categorised by age group (n=76).

Vaccination of susceptible animals with a multivalent clostridial vaccine is an important step in the control of clostridial disease on-farm.

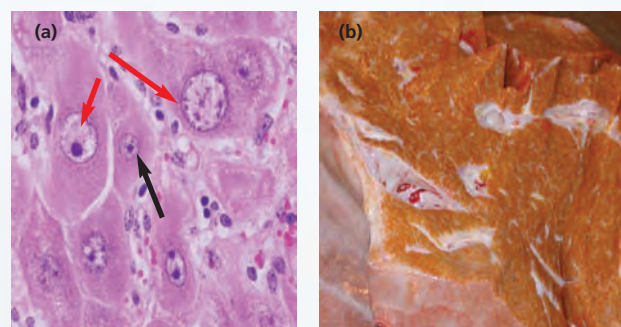
## Poisonings in Cattle

Poisoning accounted for 59 deaths in cattle in 2009. As in previous years lead was the most frequently detected cause of poisoning (Table 2).

Poisonous Agent	Neonatal	Calf	Weanling	Adult	Totals
Lead	1	6	12	7	26
Ragwort	-	-	7	8	15
Bracken fern	-	-	-	6	6
Selenium toxicity	-	-	-	3	3
Yew tree	-	-	-	3	3
Copper toxicity	-	1	-	1	2
No agent identified	-	2	-	-	2
Organo phosphorus/chlorine toxicity	-	-	-	1	1
Warfarin	-	1	-	-	1
Grand Total	1	10	19	29	59

**Table 2:** The frequency of detection of poisonous agents in cattle by age group in 2009 (n=59).

Deaths attributed to lead poisoning were distributed across all the age categories, with the weanling category (12 deaths) accounting for the majority of cases.



**Figure 25:** (a) Megalocytosis (gross enlargement of hepatocytes) associated with ragwort poisoning in the bovine liver. A normal hepatocyte is marked with a black arrow while the enlarged hepatocytes are marked with red arrows (Photo: Cosme Sánchez-Miguel). (b) Extensive hepatic fibrosis associated with ragwort poisoning (Photo: John Fagan).

Ragwort (*Senecio jacobaea*) is a hardy biennial weed which produces the characteristic yellow flowering head in its second year of growth. The pyrrolizidine alkaloids which ragwort contains are cumulative poisons. The plant is normally unpalatable for animals but becomes more palatable when it is dried in hay or ensiled. Ragwort poisoning (15 deaths) continues to be diagnosed in a significant number of cattle each year. All of the 15 cases identified in 2009 were in weanlings or adults (Figures 25 (a) and (b)).



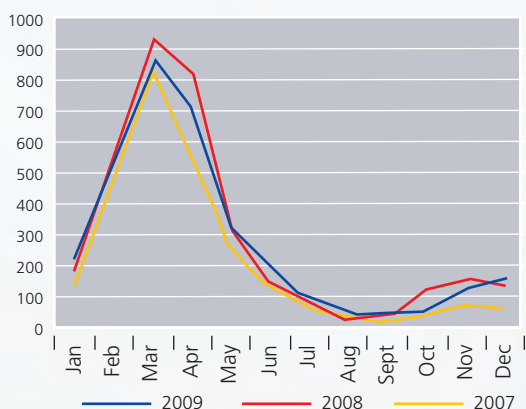
**Figure 26:** On the right is a piece of yew (*Taxus baccata*) clipping found in a field in which an adult bovine had been found dead. On the left are fragments of yew twigs, with occasional leaves, that were found in the rumen of this animal (Photo: Dónal Toolan).

Fern poisoning (six deaths) and Yew tree poisoning (three deaths) were other significant plant poisonings diagnosed (Figure 26).

Deaths of cattle attributed to mineral intoxication were less frequent, with selenium intoxication and copper intoxication diagnosed in three and two animals respectively.

## Bovine Neonatal Enteritis

Neonatal enteritis continues to be the most common cause of deaths in calves aged less than one month of age (see Figure 11).

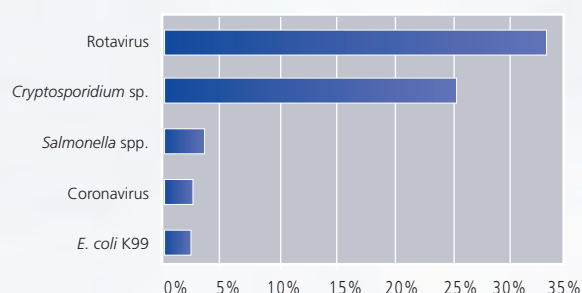


**Figure 27:** Submission numbers of faecal samples from diarrhoeic calves by month for the years 2007, 2008 and 2009.

The RVL 'calf enteritis package' comprises a series of tests performed on faecal samples from calves under one month of age to identify enteric pathogens of calves. Submission numbers for the calf enteritis package are shown in Figure 27. These follow a predictably seasonal pattern, with higher numbers of submissions received in both 2008 and 2009 than in 2007, particularly in April and May. This pattern was most probably due to adverse weather conditions in the late Spring and early Summers of 2008 and 2009 (see Figure 6), resulting in delayed turnout of animals to pasture.

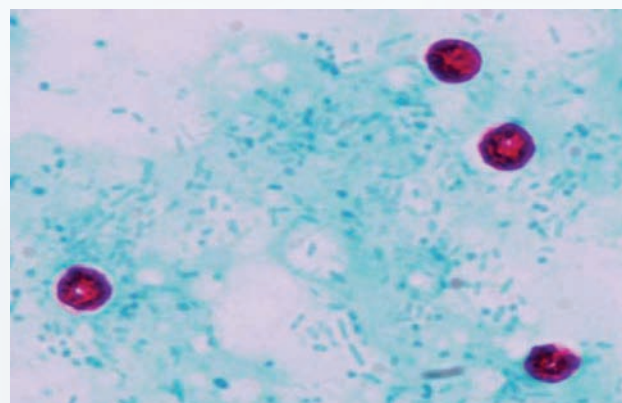
It is important that the age of calves sampled is included on the submission form which accompanies faecal samples to the laboratory. The significance of the results of the 'calf enteritis package' is dependent on the age of the calf.

The risk of enteric infection among housed calves increases as the calving season progresses – particularly if appropriate hygiene measures are not followed.



**Figure 28:** The relative frequency of calf faecal pathogens detected in samples from calves less than 1 month of age in 2009 (n=3209).

The relative frequency of identification of pathogens in calf faecal samples is shown in Figure 28. Rotavirus, at 33.1 per cent, was the most frequently identified pathogen in 2009. Cattle of all ages can carry rotavirus, and seemingly healthy animals can shed the virus. In young calves, rotavirus targets the villi in the gut wall, interfering with normal milk digestion, and leading to diarrhoea and possible death due to dehydration, acidosis and inanition.



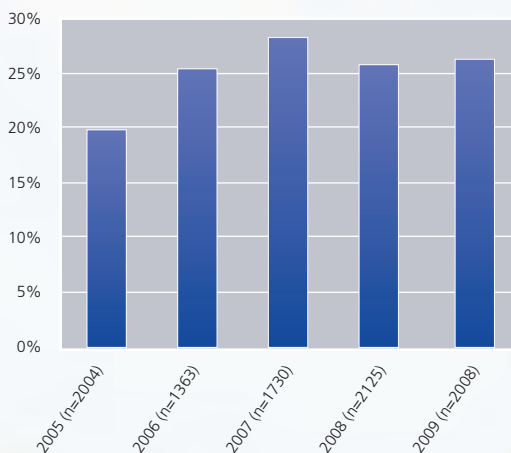
**Figure 29:** *Cryptosporidium* spp. oocysts, identified using Modified Ziehl Nielsen (MZN) stain, in the faeces of a calf (Photo: Cosme Sánchez-Miguel).

*Cryptosporidium* spp. (Figure 29) were the second most frequently identified pathogens (25.2 per cent) in neonates in 2009. Calves with cryptosporidiosis typically develop diarrhoea between seven and twenty one days of age. The agent is resistant to many disinfectants, and is fairly resistant to drying and freezing. The best approach to control is strict adherence to hygienic practices in calf housing.

Neonatal enteritis represents a significant drain on the resources of calf producers. General principles for control of neonatal enteritis include feeding sufficient colostrum, appropriate nutrition of young calves, batch-rearing, attention to hygienic measures (such as thorough cleaning and disinfection between batches) and factors that impact on calf comfort. Vaccination may also have a role to play in some instances.

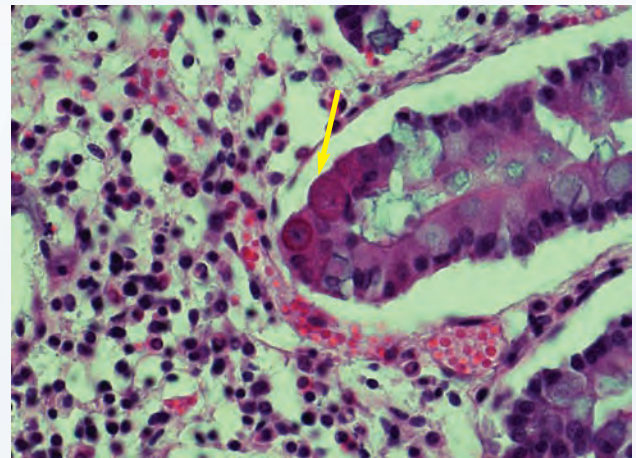
In young calves, typically those less than three days of age, *E. coli* K99 attaches to intestinal cells and causes hypersecretory diarrhoea leading to rapid dehydration and death. *E. coli* K99 was detected in 2.3 per cent of 775 faecal samples from calves less than one week of age, in 2009.

*Campylobacter jejuni* was identified in 7.9 per cent of samples from calves less than one month of age in 2009. *C. jejuni* is not usually pathogenic in cattle, but it is one of the most commonly reported bacterial enteric pathogens in humans in Ireland. The prevalence identified by the RVLs highlights the importance of animal attendants taking appropriate hygienic precautions when handling animals.



**Figure 30: Trends in the detection of *Coccidia* spp. oocysts in calf faecal samples as a percentage of all faecal samples submitted to the RVLs for *Coccidia* spp. detection in the years 2005 to 2009.**

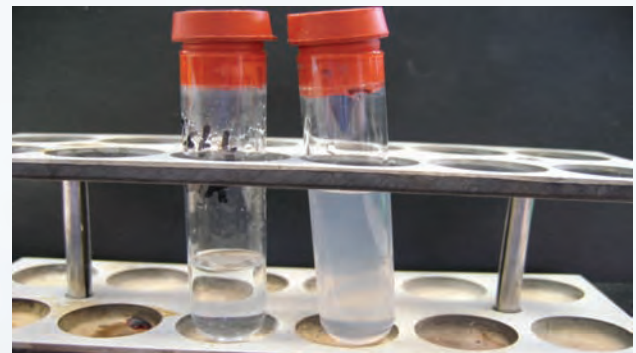
Coccidiosis is a significant cause of calf enteritis, typically in calves older than one month of age, but infection in younger calves is occasionally reported. The frequency of detection of coccidial oocysts in calf faeces in 2009 (26.4 per cent) was broadly similar to previous years (Figure 30).



**Figure 31: Photomicrograph of the intestine of a calf showing coccidial oocysts (arrow) in the enterocytes (Photo: John Fagan).**

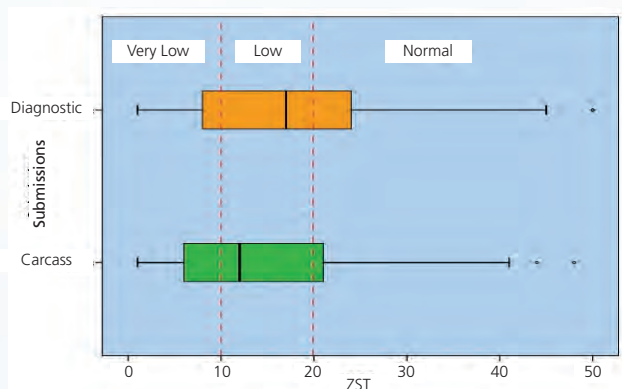
Young calves are usually infected with *Coccidia* spp. (Figure 31) when they are placed on pasture, or in housing contaminated by older cattle or other infected calves.

In calf housing, measures to improve hygiene, and in particular practices that reduce the moisture on floors, will decrease parasitic contamination as moisture favours the development and survival of the environmental stages of the parasite.



**Figure 32: The Zinc Sulphate Turbidity Test is a crude measure of antibody levels in calves up to about 10 days of age. The tube on the left has low or no precipitation, while that on the right has significant turbidity reflecting the presence of adequate immunoglobulin G (IgG) in the sample (Photo: Ger Murray).**

Adequate and timely (within the first 12 to 24 hours of life) feeding of colostrum is a vital step in the control of calf enteritis. The Zinc Sulphate Turbidity (ZST) test (Figure 32) can be performed on serum samples from calves less than 10 days of age as an indirect evaluation of the passive transfer of colostrum immunity. By quantifying the turbidity produced by the selective precipitation of the immunoglobulins (IgG) with Zinc Sulphate, the immune status of the calf can be estimated. The results are reported as units.



**Figure 33:** A box plot showing the range of ZST unit values recorded in the serum of young live calves (diagnostic submissions), and from calves presented for *post mortem* examination (carcass submissions). The boxes represent the range in which 50 per cent of the values lie. Cutoff points of 10 and 20 units are shown (n=975).

The distribution of 975 ZST test results recorded in the RVLs for 2009 is plotted in Figure 33. Two different populations are represented to emphasise the difference between serum samples procured by the private veterinary practitioner from live animals for immunoglobulin screening, and those samples taken from calves submitted for *post mortem* examination to the RVLs. While both groups are normally affected by an ongoing pathological process, the lower range of values recorded in the latter group would suggest that inadequate feeding of colostrum to these animals is central to the progression of disease and subsequent death.

Of all samples submitted for ZST testing in the RVLs, only 34 per cent were found to have values consistent with adequate passive transfer of immunity by colostrum. When calves submitted for *post mortem* are examined in isolation, this figure falls to 29 per cent. The importance of proper colostrum feeding in reducing the incidence of neonatal disease cannot be over emphasised.

## Bovine Neonatal Pancytopenia – A Novel Disease

Bovine Neonatal Pancytopenia (BNP) is a newly emerging disease syndrome of calves under one month of age. It has recently been reported in Europe and in the UK. It is variously referred to as 'Calf Haemorrhagic Disease', 'Calf Bleeding Syndrome' or 'Blood Sweating Disease', and the first cases of the condition were reported in Germany in 2007. Since then, reports of confirmed cases have slowly risen in a number of European countries. On the island of Ireland, the first confirmed case was recently diagnosed (2010) in the Republic, and cases have also been confirmed in Northern Ireland.

Typically, clinical signs in young calves include excessive bleeding from small abrasions of the skin, or from injection sites, and the passing of large thrombi in the faeces. Affected calves normally have a high temperature and become rapidly depressed. In the majority of cases, death follows within 24 to 48 hours (mortality of about 90 per cent is commonly reported). Even aggressive veterinary treatment, including blood transfusions, normally only provides a temporary respite for the affected animal.

The disease syndrome tends to affect single calves in a herd - although multiple cases have been reported in some herds in the UK and Germany. Affected calves tend to be found in larger well managed herds, with a large proportion of affected farms also keeping sheep as well as cattle. Within countries that have identified cases, a geographical pattern to the occurrence of the disease has not been identified, nor has any breed or gender association been established.

On *post mortem* examination of affected calves, significant damage to the bone marrow has been consistently reported. Severe depletion of platelets gives rise to the typical clinical signs of poor clotting and widespread bleeding (Figure 34).

While the cause of the condition is unknown, there is no evidence that it is infectious, nor that it gives rise to food safety concerns. The fact that apparently normal calves at birth can develop clinical signs rapidly after consuming colostrum has focused the attention of researchers on the possible role of colostrum in the pathogenesis of the condition. During the 24 hours that follow the birth of an animal, the intestine allows antibodies which are contained in the colostrum to pass from the gut into the blood stream. This is an important step in the transfer of immunity from the mother to her offspring, and plays a vital role in preventing disease in the newborn calf. It is possible, that in a very small percentage of calves, some of these maternal antibodies target the bone marrow cells of the calf, leading to the clinical signs of the disease. Indeed, a similar mechanism is known to cause the destruction of blood cells in some newborn foals and piglets.



**Figure 34:** Diffuse sublingual haemorrhages in a calf with Bovine Neonatal Pancytopenia (Photo: Cosme Sánchez-Miguel).

The Department of Agriculture, Fisheries and Food's Regional Veterinary Laboratories are available to assist herdowners and veterinary practitioners with the laboratory diagnosis and investigation of any suspicious cases. To this end, the Regional Veterinary Laboratories have offered to provide free *post mortem* examinations of any young calves with clinical signs typical of the condition. While the condition is rare, with minimal losses to date, the Regional Veterinary Laboratories are always vigilant to the occurrence of novel diseases, or novel presentations of existing diseases. Herdowners are asked to notify their veterinary practitioner of any young calves with apparent clotting difficulties.

### Bovine Abortion

While occasional abortions can be expected on any farm, the loss of a pregnancy can represent a significant loss to the producer - especially if it is not fully investigated and appropriate actions taken to prevent further cases.

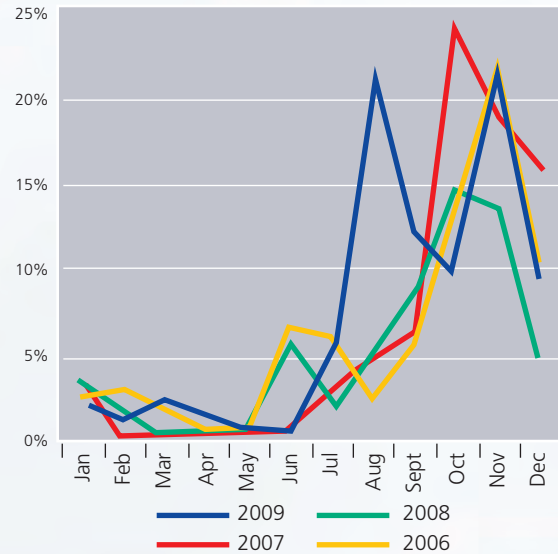
#### Infectious causes of abortion detected by culture of foetal stomach contents

Infectious agents – bacterial, fungal, protozoal and viral - represent the most commonly diagnosed causes of abortions in cattle. In 2009, 2,108 bovine abomasal content specimens were cultured.

Bacterial Agent	2006	2007	2008	2009
<i>Arcanobacterium pyogenes</i>	6.9%	5.8%	5.6%	6.7%
<i>Salmonella</i> Dublin	6.1%	7.0%	4.6%	6.0%
<i>Bacillus licheniformis</i>	4.1%	4.5%	2.7%	4.0%
<i>Listeria monocytogenes</i>	1.3%	1.6%	1.8%	1.3%
<i>Aspergillus</i> sp.	0.5%	0.9%	1.2%	1.2%
Totals	1,977	1,647	2,014	2,108

**Table 3: A comparison of bovine foetal culture results for the years 2006 to 2009<sup>5</sup>.**

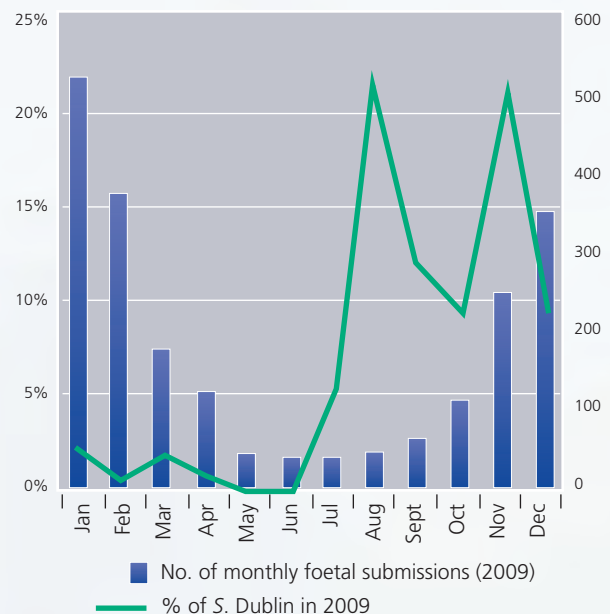
Bacteria are the most common infectious causes of bovine abortions. Table 3 shows the relative frequency of bacterial isolates from abortion submissions over the period 2006 to 2009. Except for 2007, when *Salmonella* Dublin was the most frequently identified pathogen on routine culture of foetal stomach contents, *Arcanobacterium pyogenes* has been consistently the most frequently diagnosed bacterial cause of abortion in the RVLs at 6.7 per cent. Abortion due to this organism is usually sporadic in occurrence. The bacterium reaches the placenta following bacteraemia in the cow, resulting in placentitis, and most probably taking advantage of the immature immune system of the unborn calf.



**Figure 35: The seasonal trend of *Salmonella* Dublin abortions as a percentage of all foetal submissions in the years 2006 to 2009.**

With the eradication of *Brucella abortus*, *Salmonella* Dublin has become the most common infectious cause of abortion storms in cattle (6 per cent of RVL isolates in 2009).

*Salmonella* Dublin can spread to other animals within the herd – so outbreaks of *Salmonella* abortion frequently involve multiple cases.



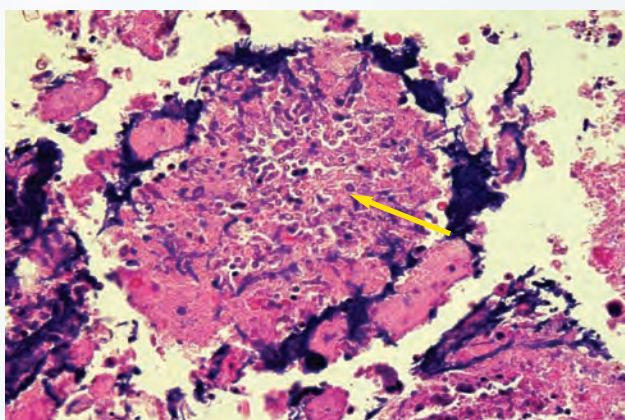
**Figure 36: *Salmonella* Dublin abortions as a percentage of all bovine foetal submissions during 2009 (n=2,108).**

Although *Salmonella* Dublin can be isolated at any time of the year, there is a seasonal increase in late autumn and early winter – see Figure 35 and Figure 36.

<sup>5</sup> Bovine foetal stomach cultures identify bacterial causes of abortion. Foetal stomach contents are the optimal sample for culture in aborted foetuses because they represent a protected sample of amniotic fluid, collected when the foetus swallowed it shortly before foetal death or expulsion, and sampled at *post mortem*.

*Bacillus licheniformis* (4 per cent) is the third most commonly detected cause of bovine abortion. This organism thrives in spoiled forage and feed, and abortions are usually sporadic. The ubiquitous nature of this pathogen makes control impractical.

*Listeria monocytogenes* was isolated from 1.3 per cent of abortion submissions. Cases are associated with high bacterial numbers of *L. monocytogenes* in poor-quality or spoiled silage. The organism gains access to the uterus and foetus via the bloodstream of the dam. This organism can also cause septicaemic and encephalitic syndromes in cattle. However, these are rarely seen in conjunction with abortions.



**Figure 37:** A bovine cotyledon with fungal hyphae of *Aspergillus* spp. (arrow) (Photo: Ger Murray).

The only fungal agents routinely identified from abortion submissions are *Aspergillus* spp. (1.2 per cent of isolates in 2009). Following dissemination in the bloodstream, *Aspergillus* spp. cause a necrotising placentitis (Figure 37). In some cases, characteristic skin lesions can be seen in the aborted foetus.

Other microorganisms isolated from foetal cultures were *Escherichia coli* (154 isolates), *Streptococcus* spp. (39), *Staphylococcus* spp. (17), *Listeria* spp. (14), *Proteus* spp. (11), *Campylobacter* spp. (5), *Pasteurella* spp. (3), *Yeast* spp. (2), *Yersinia* spp. (1) and *Nocardia* spp. (1).

While Ireland is officially brucellosis-free, selective culture for *Brucella abortus* is done as a matter of routine on abomasal contents collected from all aborted foetuses submitted to the RVLs. With the reduction in serological screening for brucellosis, this aspect of RVL surveillance work is critically important to substantiate continued disease freedom and to provide early warning of any incursion of disease.

#### Infectious causes of abortion detected by serology and other methods

Table 4 shows the frequency of detection of *Neospora caninum*, *Leptospira interrogans* serovar *hardjo*, and BVD virus, in foetal

carcasses in the years 2007, 2008 and 2009. Leptospirosis is diagnosed by the detection of *Leptospira interrogans* serovar *hardjo* specific antibodies in foetal thoracic fluid using a solid phase immunoassay method. On serological tests, titres of 1/100 or higher are considered positive. In 2009, 2.5 per cent of foetal carcasses were positive on serological tests for *L. hardjo* infection. Tissue from carcasses may also be analysed for *Leptospira* spp. using the Fluorescent Antibody Test (FAT).

Agent		2009	2008	2007
<i>L. hardjo</i> (serology)	Number Tested	521	544	431
	Percentage Positive (Titre >1/100)	2.5%	4.0%	3.2%
<i>Neospora caninum</i>	Number Tested	1282	1080	771
	Percentage Positive	4.9%	6.0%	4.9%
BVD virus (PCR/Antigen ELISA)	Number Tested	967	812	683
	Percentage Positive	5.5%	5.7%	5.6%

**Table 4:** The frequency of detection of *Leptospira hardjo*, *Neospora caninum* and BVD virus in foetal carcasses in 2007, 2008 and 2009.

*Neospora caninum* is a coccidian protozoon that causes abortion in cattle and was first identified in the 1980s. It can infect a wide variety of mammalian animals and is found world-wide. It has a two host life-cycle. Oocysts are released in dog faeces (the definitive host) and following ingestion by cattle (the intermediate host), tissue cysts are formed. During pregnancy, transplacental infection of the foetus may occur leading to either abortion or the birth of congenitally infected calves. Congenitally infected heifer calves, if retained for replacement purposes, may subsequently abort. In this way the parasite may effectively persist in herds for generations without the involvement of the definitive host. Abortion, when it occurs, is typically observed between four and six months of gestation - but early foetal loss may also occur and is more difficult to diagnose. On *post mortem* examination, neosporosis is diagnosed by the detection of *Neospora*-specific antibodies in foetal thoracic fluid. In 2009, antibodies to *Neospora caninum* were detected in 4.9 per cent of foetuses presented to the RVLs. Diagnosis may also be achieved by histopathological examination of the brain or myocardium, by immunohistochemistry (IHC), or by the Indirect Fluorescent Antibody Test (IFAT).

Bovine Virus Diarrhoea (BVD) virus infection is associated with a range of syndromes in cattle – from calf illness and death to poor reproductive performance in cows, abortions and stillbirths. BVD virus antigen was detected in 5.5 per cent of aborted bovine foetuses tested for BVD in the RVLs in 2009 (Table 4). This mirrors closely the findings recorded in 2008 and 2007.

## Bovine Mastitis

Milk samples from cows with clinical or suspected subclinical mastitis are submitted to the RVLs by veterinary practitioners for bacteriological examination to aid diagnosis, promote control, and to assist in treatment. During 2009, 5,004 milk samples were submitted to the RVLs. This represented an increase of almost 50 per cent when compared to 2008 (3,474 milk samples).

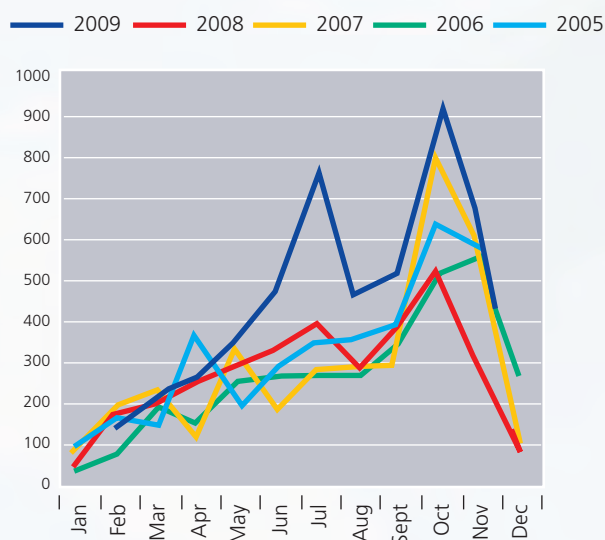


Figure 38: Milk sample submission numbers by month for the years 2005 to 2009.

The rise in milk sample submissions when compared to previous years was most apparent in July (Figure 38). It is possible that the adverse weather experienced in July 2009 (see Figure 6) which necessitated the re-housing of some stock, may have contributed to an increased incidence of mastitis in cows. Indeed, analysis of isolates from July 2009 reveals that 27.4 per cent of all *Streptococcus dysgalactiae* isolates, and 22.7 per cent of all *Streptococcus uberis* isolates cultured in 2009, were detected in this month. Both of these organisms are recognised as significant environmental pathogens - where minimising environment-to-cow transmission helps to limit their spread. Managing the environment by maintaining clean bedding, clean calving areas and clean dry teats, is imperative.

Figure 39 shows the frequency of detection of mastitis pathogens over the five years 2005 to 2009.

*Staphylococcus aureus* continues to be the most frequently isolated pathogen from milk sample submissions (43.6 per cent). The 2009 percentage, however, represents an appreciable increase when compared to 2008 (35.2 per cent), and suggests that progress has not been made in the prevention and control of this pathogen in herds.

The control of *Staph. aureus* mastitis is centred on hygienic milking procedures, proper milking machine maintenance, appropriate therapy and culling of chronic cases.

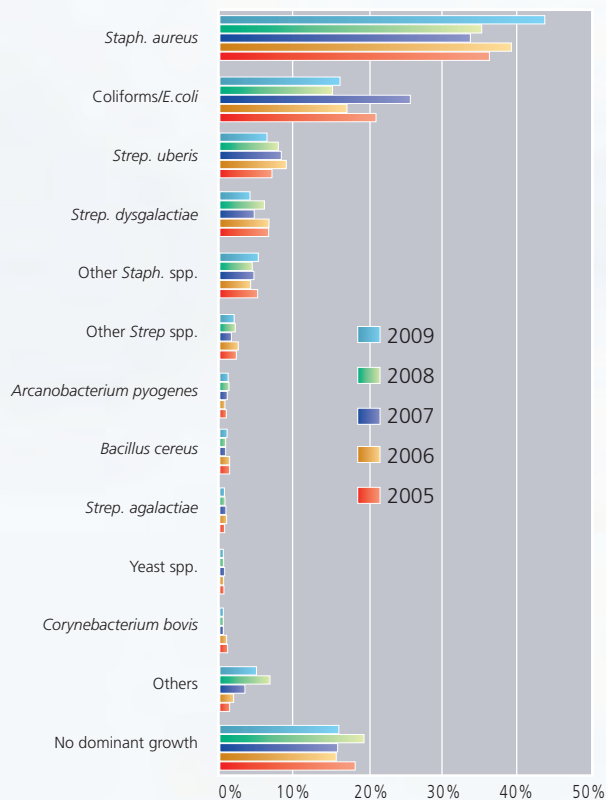


Figure 39: The relative frequency of isolation of mastitis pathogens in the years 2005 to 2009.

*Streptococcus uberis* was isolated from 6.1 per cent of milk samples analysed in the RVLs in 2009. Typically, infection with *Strep. uberis* occurs during the late dry period, when the rate of infection may be many times greater than during lactation. Cows with teat-end damage are more susceptible to infection. Flies are also thought to be involved in the spread of this bacterium from cow to cow or from the environment to cows.

*Streptococcus uberis* causes both subclinical mastitis, where the milk is normal in appearance but the somatic cell count is elevated, and clinical mastitis. The occurrence of new cases is often associated with the onset of wet and warm weather. The control of *Strep. uberis* requires proper hygienic management of the environment, particularly during the dry period and around calving.

The frequency of isolation of coliform bacteria, including *E. coli*, at 16 per cent, is predominantly attributable to the contamination of milk samples during sampling - as true coliform mastitis is generally a clinically severe condition, and is not common. Contamination of samples can be avoided by collecting aseptically into sterile containers, refrigerating immediately, and delivering the sample to the laboratory without delay.



## Bovine Respiratory Disease

Over 20 per cent of diagnoses on bovine carcasses submitted to the RVLs for *post mortem* examination in 2009 were classified as relating to respiratory disease (541 of 2,510 carcasses). This is very close to the 2008 figure of 21.5 per cent. The vast majority of the bovine respiratory disease cases are recorded as pneumonia with viral, bacterial or parasitic aetiology (Table 5 and Table 6).

	Frequency	Percentage
<i>Pasteurella multocida</i>	103	19.0%
<i>Arcanobacterium pyogenes</i>	72	13.3%
<i>Mannheimia haemolytica</i>	68	12.6%
<i>Salmonella</i> Dublin	5	0.9%
<i>Histophilus somnus</i>	3	0.6%
Various other isolates	51	9.4%
No significant growth	132	24.4%

**Table 5: Relative frequency of detection of bacterial pathogens in necropsy cases of respiratory disease (n=541)<sup>6</sup>.**

	Number Positive	Number tested	Percentage Positive
BHV1	39	375	10.4%
PI3	30	352	8.5%
Bo. Coronavirus	8	120	6.7%
BRSV	21	371	5.7%
BVDV	27	416	6.5%
<i>Mycoplasma bovis</i>	2	61	3.3%

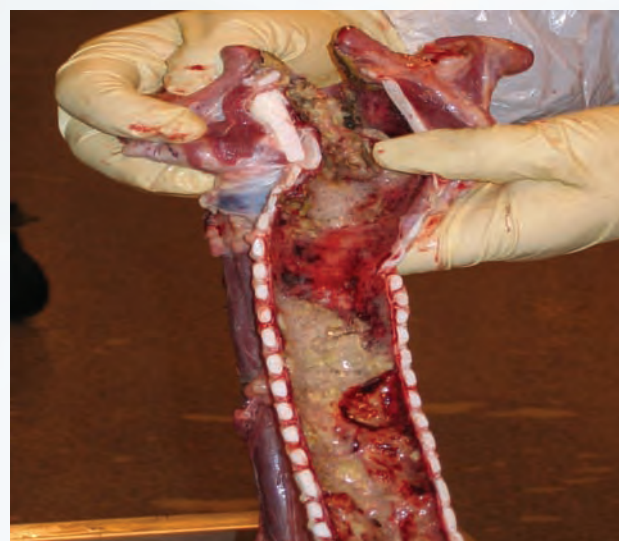
**Table 6: The relative frequency of detection of primary respiratory virus pathogens in necropsy cases of respiratory disease in 2009<sup>7</sup>.**

When respiratory disease is suspected during a *post mortem* examination, suitable pieces of affected tissue are taken for bacteriological culture. The results of these analyses are presented in Table 5. *Pasteurella multocida* was the most frequently detected bacterial pathogen - accounting for almost 20 per cent of cases. This was followed by *Arcanobacterium pyogenes* and *Mannheimia haemolytica* at around 13 per cent each. *Salmonella* Dublin was isolated from just under one per cent of lungs cultured. While *Salmonella* Dublin is not recognised as a primary respiratory disease pathogen, when pathological changes of enteritis and pneumonia are found at *post mortem* examination, it should be considered as a differential diagnosis.

<sup>6</sup> *E. coli* was the only isolate from 116 bacteriological cultures of lung tissue.

<sup>7</sup> Virus detection was by Polymerase Chain Reaction (PCR) and *Mycoplasma bovis* detection was by capture ELISA.

Bovine Herpesvirus 1 (BHV1) virus, which causes Infectious Bovine Rhinotracheitis (IBR), was the virus most frequently detected from carcasses diagnosed with respiratory disease in 2009 (Table 6). This virus was also detected in five calves less than two weeks of age - which may reflect the systemic form of the disease. Grossly, IBR is frequently associated with necrotising tracheitis (Figure 40), often exacerbated by co-infection with the bacterium *Arcanobacterium pyogenes* (10 cases).



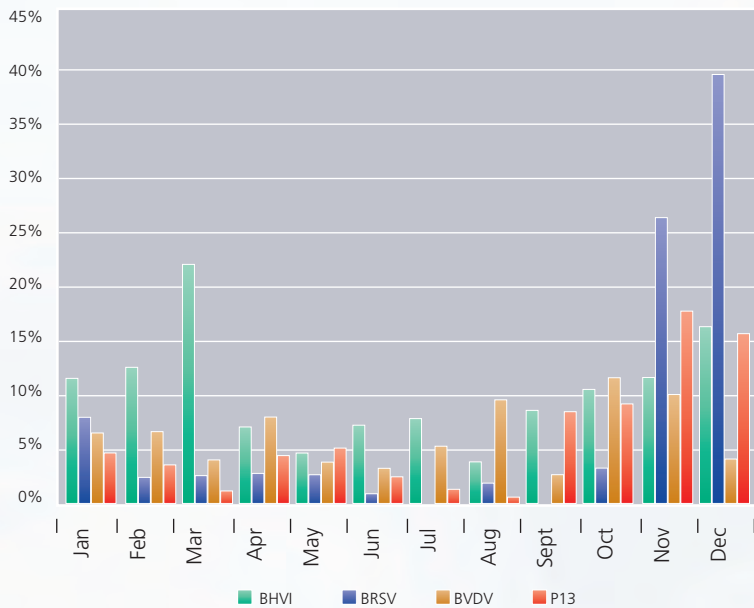
**Figure 40: Suppurative necrotic tracheitis and laryngitis in a 2 year old heifer which tested positive for IBR on Polymerase Chain Reaction (PCR) of a tracheal ring (Photo: John Fagan).**

Other bacteria found in association with BHV1 were *Pasteurella multocida* (four cases) and *Mannheimia haemolytica* (two cases).

While BVD virus is not a primary respiratory pathogen, it was identified in 14 cases. It is not possible to say whether viral detection in these cases represented transient or persistent infection. The virus was frequently detected in association with other bacterial pathogens - *Arcanobacterium pyogenes* (4) and *Pasteurella multocida* (3) being the most frequent co-infection isolates. In the carcasses in which BVD virus was the only pathogen detected, it is likely that antibiotic therapy prior to death had inhibited the growth of bacterial pathogens on routine culture. Immunosuppression attributed to BVD virus infection is often believed to play a role in bovine respiratory disease.

The frequency of detection of *Mycoplasma bovis* was low, but this pathogen requires specialised bacteriological techniques which are only carried out when the pathologist has reason to suspect its involvement.

Tuberculosis (TB) was diagnosed in nine cases of bovine respiratory disease. In addition, five cases of respiratory disease were associated with posterior vena cava thrombosis.



**Figure 41: The relative frequency of detection of viruses implicated in bovine respiratory disease during 2009 as a percentage of all nasal swabs tested by PCR at the Central Veterinary Research Laboratory (CVRL), Backweston.**

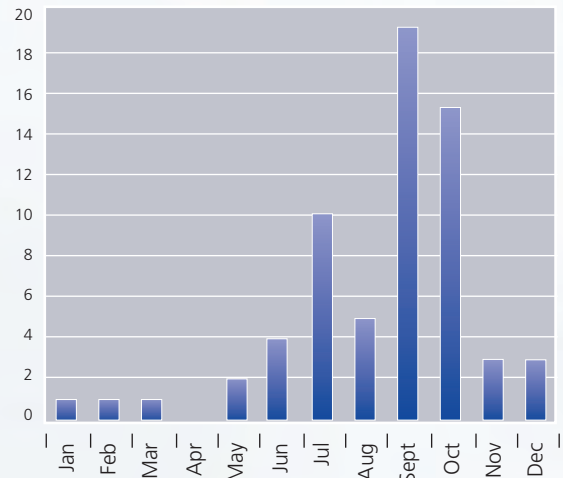
Figure 41 presents the results of respiratory virus Polymerase Chain Reaction (PCR) tests conducted in Virology Division, CVRL on tissue samples from carcasses, and on nasal swabs from acutely ill animals, during 2009. Nasal swabs are the sample of choice for the diagnosis of respiratory disease in live animals. The PCR test is an extremely sensitive method for the detection of viruses, but its success is dependent on the selection of viraemic animals for sampling, the prevention of cross-contamination, and the rapid dispatch of the samples to the laboratory.

The frequency of virus detection follows a predictably seasonal pattern - with peak detection in the winter months when animals are housed. This seasonal pattern is most notable for Bovine Respiratory Syncytial Virus (BRSV), which is the most frequently detected virus on PCR test in the winter months - while quite low detection rates are observed in the summer. December and March were the peak months for detection of BHV1.

Paired serological samples, taken during the acute and convalescent phases of disease, can also be used to aid diagnosis of respiratory disease by testing for the presence of antibodies to viruses<sup>8</sup>.

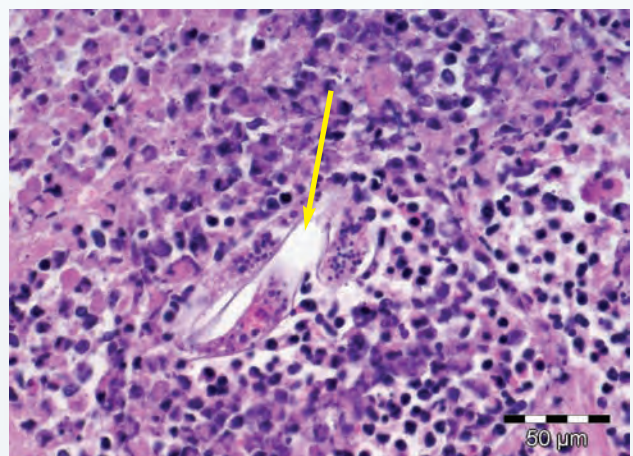
It is important that the vaccination status of animals sampled is provided on the submission form which accompanies the samples to the laboratory. This can influence the interpretation of the results.

<sup>8</sup> Owing to the high prevalence of seropositivity in cattle to both BRSV and P13 (over 95 per cent), routine serological testing to detect BRSV or P13 specific antibodies has been discontinued. Should a disease investigation warrant P13 or BRSV serology, arrangements should be made with the RVL prior to the sampling of animals.



**Figure 42: The relative frequency of diagnosis of bovine parasitic pneumonia by month in 2009 (n= 63).**

The number of parasitic pneumonia cases diagnosed in 2009 showed a substantial increase from previous years, reflecting the high rainfall during the grazing season. Sixty three diagnoses of parasitic pneumonia were recorded (representing 11.6 per cent of respiratory disease diagnoses). Numbers peaked in September and October. One case was also recorded in January, and three in December, months that would not normally be associated with parasitic pneumonia (Figure 42). The majority of the diagnoses were in the 'three to twelve months' of age category (36), while eight were in calves between one and three months of age, and seven were in animals over 12 months of age. The youngest affected animal was 10 weeks of age, while the oldest was 20 months.



**Figure 43: Photomicrograph of lungworm larvae (arrow) in the lung of a weanling (Photo: Jim O Donovan).**

Parasitic pneumonia (Figure 43) must also be considered as a differential diagnosis in cattle showing respiratory signs during their second grazing season. Some prophylactic anthelmintic regimes do not facilitate the development of immunity in the first grazing season, hence the naivety of some cattle to infection during their second season at grass.

## Johne's Disease 2009

Johne's Disease, caused by infection with *Mycobacterium avium* subsp. *paratuberculosis* (MAP), is a Class B notifiable disease. It is characterised by chronic diarrhoea, loss of thrive and eventual death (Figure 44). It is primarily a disease of ruminants, but has a wide host range.



**Figure 44: A cow with weight loss and chronic intractable diarrhoea which proved positive on faecal culture for *Mycobacterium avium* subsp. *paratuberculosis* (Photo: Ger Murray).**

Cattle typically acquire the infection as calves, either through the ingestion of colostrum or milk containing MAP, or by exposure to MAP in faeces or contaminated feed and water. *In-utero* infection of the foetus can also occur - and is considered to be of significance in herds where the prevalence is greater than 5 per cent.

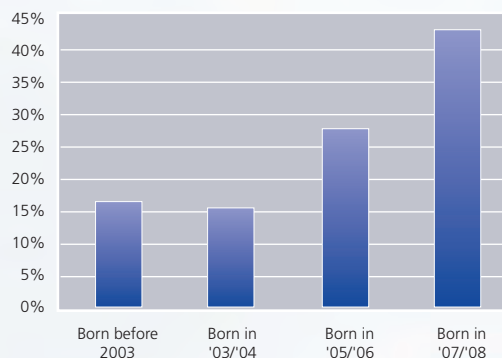
Year	Tests	Positive	Positive (%)
2005	206	35	17.0
2006	170	43	25.3
2007	304	59	19.4
2008	416	92	22.1
2009	376	103	27.4

**Table 7: *Mycobacterium avium* subsp. *paratuberculosis* (MAP) culture results from cattle for the years 2005 to 2009.**

Faecal culture is considered the 'gold standard' test for the diagnosis of Johne's Disease. It is provided by the TB Section in the Central Veterinary Research Laboratory (CVRL), Backweston, via the Regional Veterinary Laboratories.

Three hundred and seventy six samples were cultured for the presence of *Mycobacterium avium* subsp. *paratuberculosis* (MAP) in 2009. One hundred and three (27.4 per cent) were positive (Table 7). This was the highest prevalence recorded on faecal culture in the last five years. The positive animals originated in 48 herds. All except one animal were bred in Ireland - and there was a relatively even distribution between beef (56 per cent) and dairy breeds (44 per cent). Ten of the positive animals were bulls, all from separate herds and all were of the Limousin breed.

A survival analysis of positive animals, conducted by interrogation of the Animal Identification and Movement (AIM) system, shows that many were dead within six months of a positive diagnosis.



**Figure 45: An age analysis of cattle which were positive for *Mycobacterium avium* subsp. *paratuberculosis* (MAP) on faecal culture in 2009 (n=100).**

While calves typically acquire infection at less than six months of age, the development of the disease is slow and clinical disease is normally only observed in animals between two and six years of age.



**Figure 46: Corrugated folds in the ileum of a cow associated with granulomatous enteritis caused by Johne's Disease (Photo: Cosme Sánchez-Miguel).**

Clinical signs are observed in animals following the development of granulomatous enteritis (Figure 46) leading to malabsorption, a protein-losing enteropathy and intractable diarrhoea. In 2009, 47 per cent of animals which were positive on faecal culture analysis were aged less than three years.

Year	Tests	Positive	Positive (%)
2005	2001	152	7.6
2006	2185	183	8.4
2007	2755	173	6.3
2008	3372	229	6.8
2009	3981	251	6.3

**Table 8: The percentage of sera which tested positive in the *Mycobacterium avium* subsp. *paratuberculosis* ELISA for the years 2005 to 2009.**

There were 3,981 sera tested in the MAP ELISA in 2009; 6.3 per cent of these were positive. The number of samples submitted in 2009 was almost twice that received in 2005 (Table 8). It should be noted that false-positive serological results can arise due to infection with non-pathogenic environmental mycobacteria. Tuberculin testing may also cause certain animals to have false positive responses in the MAP ELISA during the two month period after intradermal testing. The true infection status of a live animal can only be established through the use of faecal culture.

## Biosecurity

In any discussion of disease prevention, farm biosecurity plays a vital role. Biosecurity should be part of the general farm management to limit incursion of infectious disease or the spread of endemic disease on the farm.



**Figure 47: The use of (a&b) appropriate signage and (c) footbaths at the entry points to farms are simple and effective biosecurity measures to prevent the transmission of disease (Photos: Michael Gormley).**

The herd veterinarian has a critical role in advising on biosecurity. Specifically, such advice refers to controlling the movement of people (Figure 47 (a) & (b)) and animals onto and within the farm, grouping of animals according to their age, and adherence to effective vaccination protocols.

Protection of the herd or flock begins by preventing the introduction of infectious disease. This can be achieved by maintaining adequate farm boundaries, operating a closed herd, and the quarantining and testing of purchased animals before their introduction to the herd. The importance of appropriate disinfection of protective footwear at the entry point to the farm (Figure 47(c)), as well as the disinfection of transport vehicles should not be overlooked.

The control of endemic disease on farm requires measures such as batch rearing animals with appropriate disinfection between batches, isolation and prompt treatment of sick animals, and the separation of age-groups while housed. Further measures such as maintaining clean water and feed troughs, rodent control and preventing dog or cat access to feed stores are equally important.

Additional biosecurity measures which should be adhered to include:

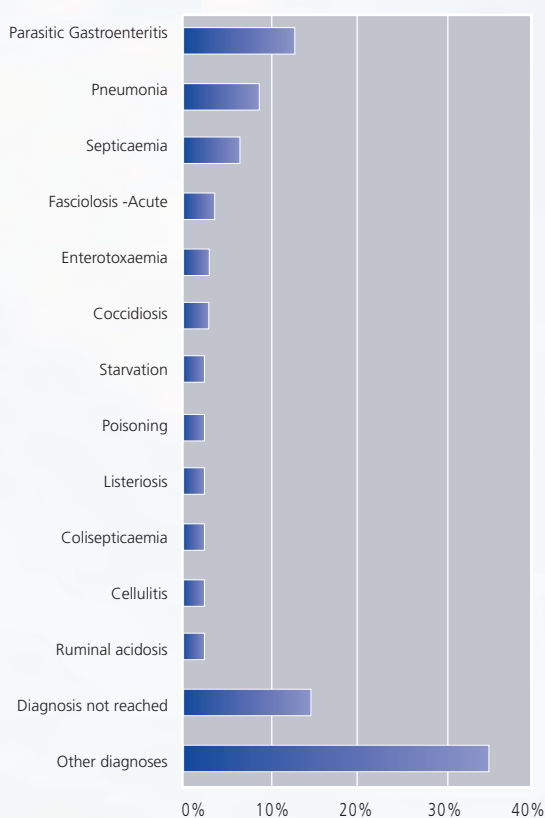
- Try to purchase cattle from herds with a known herd status.
- Avoid purchasing cull cows.
- Quarantine animals returned unsold from the mart for 3 weeks before reintroducing them to the herd.
- Do not use calving pens for sick animals
- Disinfect equipment before use – this is especially important with stomach tubes. Never use a stomach tube for colostrum feeding where it has been used on sick calves.

The implementation of effective biosecurity measures promotes animal health and welfare and consequentially farm productivity. Further details can be accessed at <http://www.agriculture.gov.ie/animalhealthwelfare/diseasecontrol/>

## Diseases of Sheep

### Mortality in lambs (birth to six months)

The number of submissions of lambs less than six months of age fell by 22.8 per cent to 220 in 2009, reversing the previous year's increase in submissions in this category. The most frequent diagnoses in young lambs in 2009 were parasitic gastroenteritis (12.7 per cent) and pneumonia (8.6 per cent) – see Figure 48.



**Figure 48: The relative frequency of the diagnosed causes of deaths of lambs under six months of age in 2009 (n = 220)<sup>9</sup>.**

Septicaemia (6.4 per cent) was the next most frequent diagnosis. As with pneumonia in this age group, the agent most frequently associated with septicaemia was *Mannheimia haemolytica*. The heavy contamination of pasture with liver fluke eggs (*Fasciola hepatica*) in 2009 is reflected in the diagnosis of acute fasciolosis as the cause of death of 3.2 per cent of lambs less than six months of age. This is an exceptional finding in lambs of this age, and has implications for future fluke control programmes on affected farms.

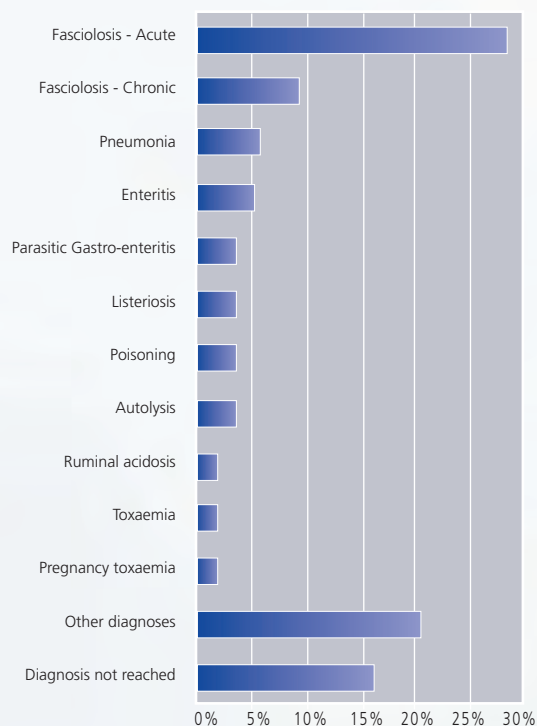
<sup>9</sup> In this age group, the category "Diagnosis not reached" includes lambs which were unsuitable for examination due to autolysis.

It is likely that diagnoses of colostrum deprivation and hypogammaglobulinaemia are under-represented in these figures, as deaths of colostrum-deprived neonatal lambs are often recorded as other diagnoses such as septicaemia or pneumonia. Low maternally-derived antibody levels continued to be an underlying factor in the epidemiology of all neonatal lamb mortality in 2009.

Diagnoses associated with the deaths of less than 1.5 per cent of lambs examined have been grouped under the heading of 'Other diagnoses'. This category includes small numbers of cases of nephrosis, intestinal obstruction, and malignant oedema, as well as other less frequent diagnoses.

### Mortality in adult sheep (over six months)

In contrast to the number of young lambs submitted to the RVLs for *post mortem* examination, the number of submissions of adult sheep increased by 50.6 per cent in 2009. This is despite a further contraction in the size of the national flock (see Figure 4).



**Figure 49: The relative frequency of diagnosed causes of deaths of adult sheep carcasses examined in 2009 (n = 396).**

Acute fasciolosis (28.3 per cent) was the most frequent diagnosis in adult sheep (Figure 49). Acute and chronic fasciolosis cases combined, accounted for 37.4 per cent of all adult sheep deaths diagnosed in the RVLs in 2009. This was almost double what was considered to be a high figure of 20.5 per cent in 2008.

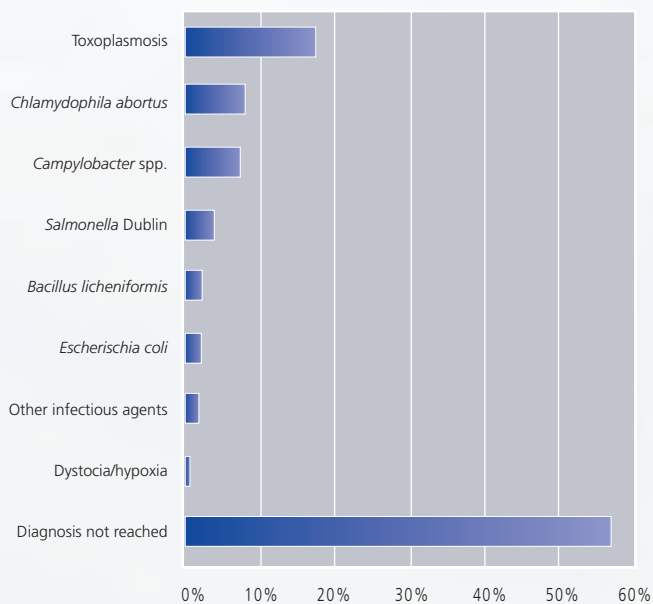


**Figure 50: A pale and friable liver from a ewe with immature liver fluke tracts caused by migrating *Fasciola hepatica* (Photo: Cosme Sánchez-Miguel).**

As in 2008, pneumonia (5.3 per cent), enteritis (5.1 per cent) and parasitic gastroenteritis (3.5 per cent) were significant causes of mortality in adults. Seventeen diagnoses of listeriosis (2.8 per cent) reflected the difficult silage harvesting conditions of the previous year – resulting in a higher proportion of poor silages, which provide optimum conditions for listerial proliferation.

## Ovine Abortion

Determination of the cause of ovine abortion is an important step in preventing further perinatal losses in an affected flock. Specimens from cases of ovine abortion are submitted to the RVLs to determine the cause, and in particular to rule out specific infectious causes of abortion - as these could be mitigated by taking preventive action such as vaccination.



**Figure 51: The relative frequency of different causes of ovine abortion in 2009 (n = 175).**

There were 175 ovine abortion submissions to the RVLs during 2009 - comprising 303 foetuses. A submission from a case of ovine abortion usually consists of more than one aborted foetus and may also include foetal membranes. The relative frequencies of diagnoses for such submissions to the RVLs in 2009 are shown in Figure 51. In a pattern similar to that observed in recent years, toxoplasmosis (17 per cent) was the most frequently diagnosed cause of abortion in ewes in 2009 - followed by enzootic abortion of ewes (EAE) caused by *Chlamydomphila abortus* (8 per cent).

*Chlamydomphila abortus* infection is spread at lambing time - uninfected female sheep acquire infection from the foetal membranes and bedding contaminated by fluids from aborted ewes. The infection then becomes inactive, reactivating during the middle of the next pregnancy. This allows EAE to persist in flocks from year to year.

*Chlamydomphila abortus* and *Toxoplasma gondii* are both zoonotic pathogens which can pose a risk to the unborn child. Pregnant women should avoid all contact with sheep, especially at lambing time.



**Figure 52: Lesions in a foetal liver highly suggestive of intrauterine infection with *Campylobacter* spp. (Photo: Dónal Sammin).**

Abortions associated with *Campylobacter* spp. (Figure 52) were more frequently detected in 2009 (seven per cent of diagnosed cases) than in the previous two years (approximately three per cent of diagnosed cases). *Campylobacteriosis* in ewes is transmitted orally and is very contagious in housed ewes where animals are in close proximity. Typically, it causes abortion in the last six weeks of pregnancy. Premature lambing, stillbirths, diarrhoea and vaginal discharge before parturition may also be noted. Infected animals acquire limited immunity, and a ewe that aborts usually will not do so again for some time. As aborted tissue is infectious to sheep and other animals, all aborted tissues, foetal membranes, and discharges should be removed to prevent the spread of infection, and appropriate disinfection measures should be undertaken.

A definitive diagnosis was not reached in a relatively high proportion of abortion submissions (57 per cent). This is consistent with international norms. In many instances this is due to the unsuitability of specimens for diagnosis (advanced autolysis of tissues), inadequate sample submission (i.e. the submission of only a single foetus), or failure to submit the foetal membranes. Non-infectious sporadic causes of abortion, such as physical trauma or environmental or nutritional stresses, are also much less likely to be identified in the laboratory than infectious causes.

The category 'Other infectious agents' in Figure 51 includes abortions attributed to *Listeria monocytogenes* (n = 1), *Yersinia pseudotuberculosis* (n = 1) and an *Aspergillus* sp. (n = 1).

Submission of foetal membranes from aborting ewes is critical for optimal laboratory diagnosis of enzootic abortion. *Chlamydophila abortus* is most readily detectable in placental tissue. The diagnostic pathological change associated with this infection is severe suppurative inflammation of the foetal membranes (Figure 53).



**Figure 53: Foetal membranes from a ewe exhibiting suppurative inflammation caused by *Chlamydophila abortus* (EAE) infection (Photo Jim O Donovan).**

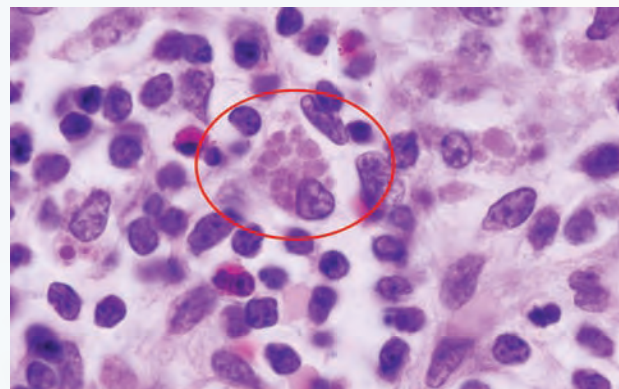
In cases where foetal tissues may not be available for submission, but where a significant proportion of a flock have aborted, blood should be collected from a number of aborted ewes and submitted for antibody testing. High antibody levels to infectious agents (i.e. *Toxoplasma gondii* or *Chlamydophila abortus*) are evidence of exposure. In some cases, comparison of antibody levels from a number of ewes that have lambed normally in the same flock may assist in interpretation.

## Diseases of Pigs

### Postweaning Multisystemic Wasting Syndrome (PMWS)

First described in 1991, PMWS is consistently associated with a porcine circovirus (PCV), but development of the disease is most notable in animals with concurrent infection with other viruses. Consequently, only a proportion of seropositive pigs develop the clinical disease. There are two serotypes, Type 1 which is not known to cause disease and Type 2 which can be isolated from PMWS lesions. There are several different strains.

The most typical clinical sign is a progressive loss of body condition. Lymph nodes are routinely enlarged and dyspnoea, pallour of the skin, jaundice, and sometimes diarrhoea, may be noted. PMWS may delay finishing in pig herds, resulting in significant economic loss. Mortality in excess of 50 per cent is often recorded. Immunosuppression probably plays a significant role in the development of the disease.



**Figure 54: Lymph node from a pig with PCV2 botryoid intracytoplasmic inclusion bodies in a histiocyte (Photo: Cosme Sánchez-Miguel).**

The virus is found in blood, saliva, faeces, urine, and semen of infected pigs and transmission is primarily by direct contact. The PCV2 virus is quite resilient, allowing it to survive in the environment if rigorous sanitary measures are not adhered to between pig batches.

Typical *post mortem* findings include pneumonia (pulmonary oedema and consolidation), jaundice of the liver, and enlarged kidneys with white foci on the surface. Myocarditis is sometimes noted in chronically infected pigs. Diagnosis of PMWS relies on the identification of the PCV2 virus, together with associated clinical signs and pathological changes (Figure 54). In addition to routine *post mortem* examination, the Veterinary Laboratory Service provides PCV2 serology and immunohistochemistry to aid diagnosis in pigs with clinical signs of PMWS. PMWS was diagnosed in 3.9 per cent of pig carcasses submitted to the RVLs in the last four years.

## Accidental Selenium Intoxication in a Pig Herd

In the winter of 2009, Cork RVL investigated unexpected sudden deaths and illness on two pig farms. The farmer and private veterinary practitioner noted a marked increase in the demand for water by both sows and gilts. This was followed by a decrease in appetite and feed intake, loss of body condition, disruption in the normal service patterns, some abortions in the final third of pregnancy, and premature farrowing. Some of the sows collapsed and died suddenly. Remarkably, suckling and weaned piglets were unaffected. During the outbreak, more than 20 of the 500 sows died and many more were seriously affected.

Gross examination and routine tests were unremarkable; however, analysis of kidney and liver tissues identified a moderate to high concentration of selenium in many of the animals examined<sup>10</sup>. The ration which was fed to the pigs was also analysed<sup>11</sup> and was found to have a higher concentration of selenium than normal.



**Figure 55: Sows' feet showing hoof separation, haemorrhages, deformed hoof walls and horizontal fissures and cracks (arrows), equidistant from the coronary band (Photo: Cosme Sánchez-Miguel).**

Accidental selenium intoxication is rare, but when it occurs it is normally observed as an acute syndrome characterised by ataxia and hind limb paralysis caused by focal, bilaterally symmetrical poliomyelomalacia in the ventral horns of the spinal cord. Severe neurological signs were not a feature of this outbreak, and the characteristic histological lesions were not seen in any of the animals examined *post mortem* in the RVL. This may have been because the animals involved were not exposed to extremely high levels of selenium. However, subacute selenium intoxication, targeting the integument, became more evident during a subsequent visit to the farm when most of the surviving sows displayed hoof separation and deformed hoof walls, with horizontal fissures and cracks, equidistant from the coronary band in each affected foot (Figure 55).

In a related incident on a second pig farm, 130 out of 959 weaners died over a three day period after the introduction of a new batch of feed, which had been accidentally contaminated with high levels of selenium. Acute selenium intoxication was diagnosed on the basis of high selenium concentration in kidney, liver, blood and feed samples.

## Swine Influenza in a Pig Herd

Influenza A viruses are highly infectious respiratory pathogens that have a wide host range. Swine Influenza is a scheduled and notifiable disease in Ireland. Clinically it is associated with the sudden onset of fever, depression, coughing, ocular and nasal discharge, and abortion. Various strains of the virus exist, of which two have been isolated in Ireland - H1N1 (1991), and H3N2 (1993). The H1N1 that has circulated in Ireland since 1991 is different from the strains circulating in Europe, and is possibly the product of the introduction of an avian strain into Irish pigs. Vaccination for swine influenza is not routinely practiced in pigs in Ireland. The pandemic strain of H1N1 (2009) was detected for the first time in twelve sows in a farm in the south of the country on 29 September 2009. Samples were taken and submitted to Virology Division, CVRL, Backweston, where Avian Influenza H1N1 was confirmed. The source of the virus was not identified, but it was most likely introduced into the pigs *via* an infected person, or by the movement of pigs from an infected herd.

Swine are susceptible to infection with both avian and human influenza viruses. This can allow novel re-assortant influenza viruses to be generated when pigs are concurrently infected with avian and human strains. For this reason, vigilance on the part of herdowners and veterinarians is paramount for the identification of clinical signs of swine influenza, as well as the strict adherence to appropriate biosecurity measures on-farm.

<sup>10</sup> Tissue and blood selenium analyses were performed in the Clinical Chemistry Section, Backweston.

<sup>11</sup> Feed analysis was performed in the State Laboratory, Backweston.



## Parasitic Diseases

The number of faecal samples submitted to all RVLs for parasitological examinations increased dramatically in 2009. This may be due to the fact that the risk of parasitological infections was considered to be high for most of the year due to weather conditions, and at the same time the need for vigilance had been widely publicised. Figure 56 shows the marked increase in bovine faecal submissions to the RVLs for fluke egg examination over the years 2006 to 2009.

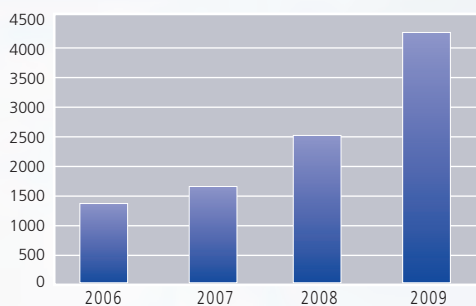


Figure 56: The number of bovine faecal samples submitted for fluke egg detection in the years 2006 to 2009.

## Liver Fluke and Rumen Fluke Infections

In 2009, the percentage of bovine and ovine faecal samples positive for *Fasciola hepatica* eggs was higher than seen in recent years. Figure 57 illustrates the monthly trends in detection rates in bovine samples, with a peak of 34.2 per cent of samples positive for *Fasciola hepatica* eggs in May. Three of the most significant factors associated with outbreaks of fasciolosis (the ambient temperature, moisture levels in the local environment, and the availability of suitable snail habitats) were facilitated by the wet and mild winter of 2008-2009, and the wet summer of 2009 (see 'Weather in 2009' on page 4).

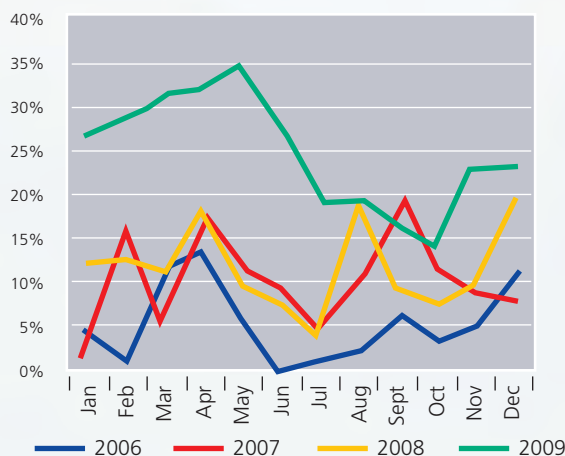


Figure 57: A comparison, by month, of the percentage of bovine faecal samples positive for liver fluke eggs in the years 2006 to 2009.

The number of cases of severe acute fasciolosis in sheep, diagnosed following *post mortem* examinations in the RVLs, reached levels in the autumn and winter of 2009 which have not been seen in recent years (see Figure 48 and Figure 49). Of 795 ovine faecal samples examined for the presence of liver fluke eggs during 2009, two hundred and twenty five (28.3 per cent) were positive, which represents a significant increase when compared to 2008 (13 per cent) and 2007 (4.5 per cent). Figure 58 compares the trends in the detection rates of *Fasciola hepatica* eggs in ovine faeces by month during 2008 and 2009.

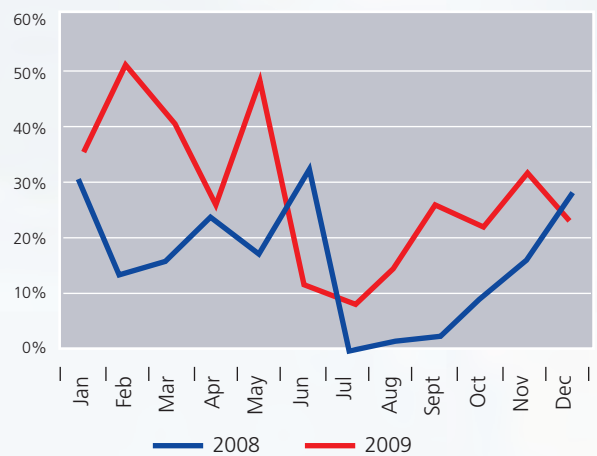


Figure 58: A comparison, by month, of the percentage of ovine faecal samples positive for liver fluke eggs in the years 2008 and 2009.

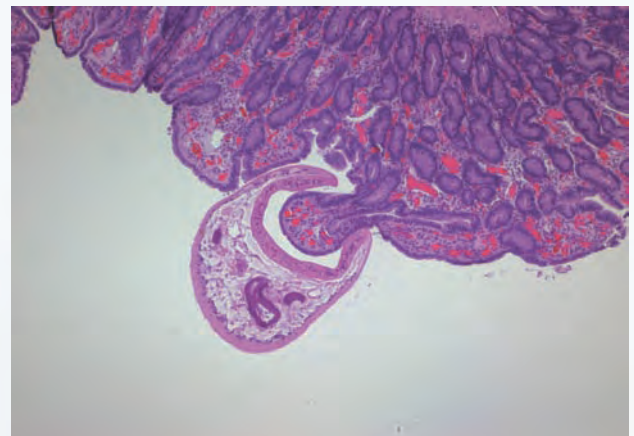


Figure 59: Photomicrograph showing *Paramphistomum* spp. larva attached to the mucosa of the small intestine (Photo: Dónal Sammin).

From August 2009, the RVLs began reporting on the presence or absence of rumen fluke eggs (*Paramphistomum* spp.) in faecal samples.

It had been noted in recent years that rumen flukes had been seen more frequently during *post mortem* examinations. The presence of rumen fluke eggs in the faeces of an animal only indicates that adult flukes are present in the forestomachs, and does not confirm their involvement in clinical disease.

In the face of an acute disease episode (acute larval paramphistomosis), which is typically associated with immature larvae in the upper small intestine (Figure 59), faecal analysis is of limited use in diagnosis, as the parasite is still in the pre-patent (non-egg laying) phase. Rumen fluke eggs are quite similar in size and shape to liver fluke eggs, but can be differentiated by the fact that they are clear rather than yellow in colour (Figure 60).

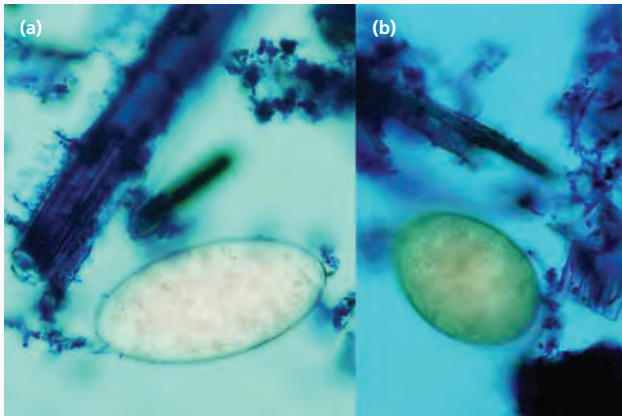
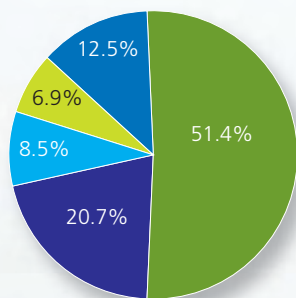


Figure 60: Photographs of (a) a rumen fluke (*Paramphistomum* sp.) egg and (b) a liver fluke (*Fasciola hepatica*) egg. Rumen fluke eggs are larger and paler than liver fluke eggs (Photo: Cosme Sánchez-Miguel).

### Gastro-intestinal Parasitic Infections

Eight hundred and eighty one ovine faecal samples were examined for the presence of strongyle eggs during 2009. Eggs were not detected in 51.4 per cent of the samples. In 12.5 per cent of samples, very high strongyle egg numbers (>1600 eggs per gram) were detected. These figures were similar to 2007 and 2008. Figure 61 summarises the 2009 findings, and categorises the infection according to the number of eggs recorded on faecal examination.



- Negative
- 100-400epg (Low)
- 500-800epg (Moderate)
- 900-1600epg (High)
- >1600epg (Very High)

Figure 61: The percentage of ovine faecal samples positive for strongyle eggs in 2009 (n = 881) (epg = eggs per gram).

*Nematodirus* spp. eggs (Figure 62) are about twice the size of the typical strongyle egg and can be identified quite easily on microscopic examination.



Figure 62: A photomicrograph of a *Nematodirus battus* egg (Photo: Brendan Crowe).

Nematodiriosis can be a significant cause of diarrhoea in sheep, particularly in young lambs. Development to the L<sub>3</sub> larval stage takes place within the egg, and in the case of *Nematodirus battus* (the most significant species seen in Ireland), a prolonged cold period is required before hatching from the egg occurs. It is common therefore that large numbers of L<sub>3</sub> larvae appear on infected pastures in April and May when young suckling lambs are beginning to ingest more grass. If young naive lambs ingest enough of these larvae, severe clinical disease can result.

Ten per cent of ovine faecal samples examined for strongyle eggs in 2009 were positive for *Nematodirus* spp. – with the highest proportion of positive samples in May (25 per cent of samples examined). Figure 63 gives a comparison of the monthly trends in detection rates over the years 2007 to 2009.

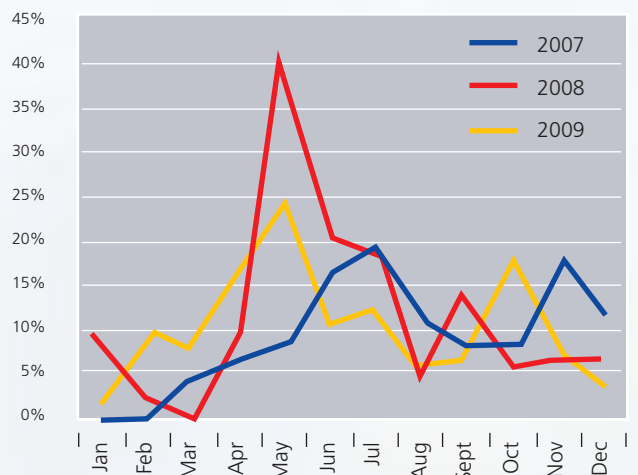
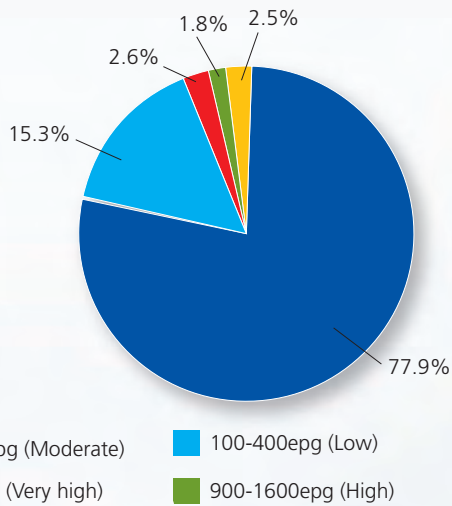


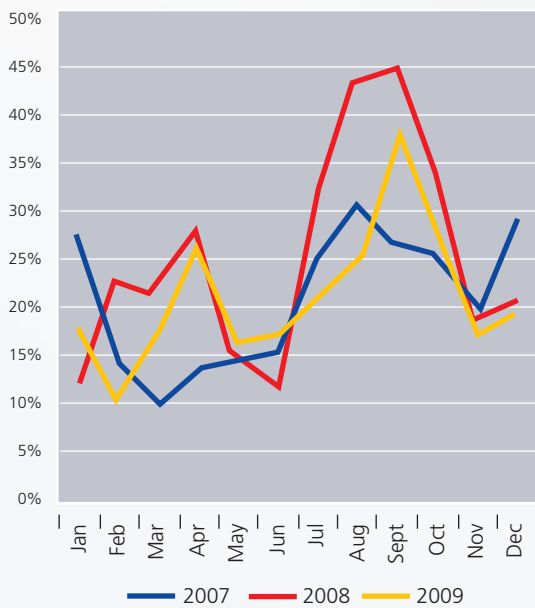
Figure 63: A comparison, by month, of the percentage of ovine faecal samples positive for *Nematodirus* spp. eggs in the years 2007 to 2009.

A total of 4,665 bovine faecal samples was examined for the presence of strongyle eggs in 2009 - an increase from 3,330 in 2008. In 77.9 per cent of the samples, no strongyle eggs were detected. In 2.5 per cent of samples, very high strongyle egg numbers (>1600 eggs per gram) were detected. Figure 64 summarises the results for the year, and categorises infections according to the number of eggs recorded on faecal examination.



**Figure 64:** The percentage of bovine faecal samples positive for strongyle eggs in 2009 (n = 4,665) (epg = eggs per gram).

Figure 65 illustrates the frequency of significant strongyle egg detection (>400 eggs per gram) in bovine faecal samples by month over the years 2007 to 2009.



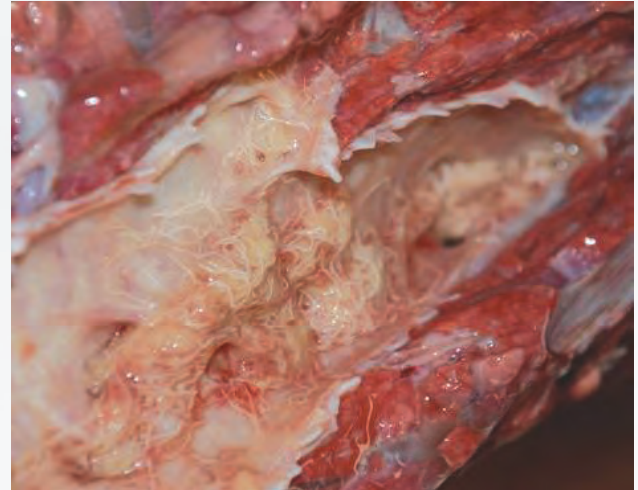
**Figure 65:** The percentage, by month, of bovine faecal samples with significant strongyle egg counts (>400 eggs per gram) in the years 2007 to 2009.

### Lungworm Infections

Year	No. Tested	No. Positive	% Positive
2007	566	25	4.4%
2008	805	29	3.6%
2009	1203	70	5.8%

**Table 9:** The percentage of bovine faecal samples which were positive for the presence of lungworm larvae (primarily *Dictyocaulus viviparus*) in the years 2007 to 2009.

In 2009, 1,203 bovine faecal samples were submitted to the RVLs for examination for the presence of lungworm larvae (primarily *Dictyocaulus viviparus* – see Figure 66). Both submission numbers and the percentages positive were higher in 2009 than in either of the two previous years (Table 9). Further data on lungworm diagnoses in bovine carcasses submitted to the RVLs is presented on page 17.



**Figure 66:** Tangled masses of adult lungworm in the trachea of a yearling bullock (Photo: Cosmé Sanchez-Miguel).

### The *Trichinella* spp. Survey 2009

*Trichinella* species (almost exclusively *Trichinella spiralis*) are nematode parasites that are significant internationally as zoonotic agents, with pigs and humans being the most important hosts. The parasite cycles at a low level in wild animals, especially rodents, which act as a reservoir with the potential to infect pigs. Foxes are used for surveillance as they are a top predator and provide an efficient and accurate estimate of the prevalence in the wild mammalian population.

A risk-based *Trichinella* monitoring programme in wildlife has been established in Ireland, a country where the risk of trichinellosis in domestic pigs is officially recognised as negligible. The RVLs carry out this work in compliance with Commission Regulation (EC) No. 2075/2005, which lays down specific rules on official controls for *Trichinella* in meat intended for human consumption.

In 2009, 442 foxes were collected from multiple locations across the Republic of Ireland. Specified samples of skeletal muscle from each fox were examined in the RVLs. Three foxes (one each in Monaghan, Cork and Limerick) were found to be positive for *Trichinella* spp. larvae. Figure 67 shows the locations of origin of all the foxes tested.

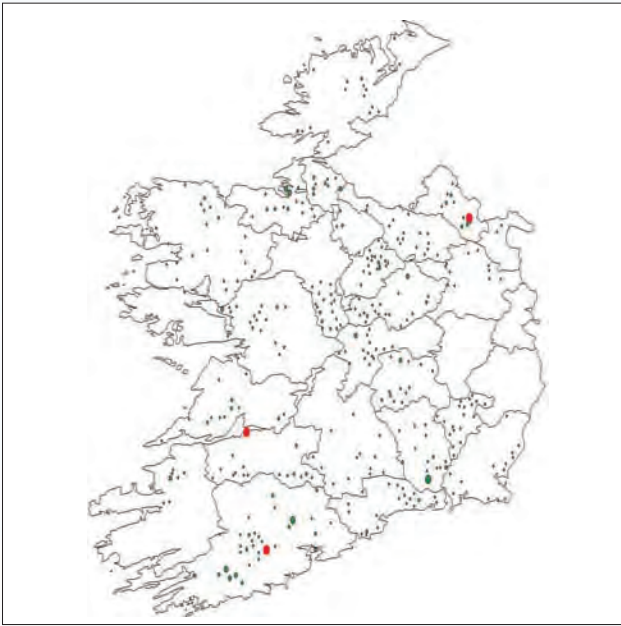


Figure 67: Locations where foxes were collected for the *Trichinella* spp. survey in 2009 (n = 442). Larger dots represent locations where more than one fox was collected. The sites where the positive foxes were located are marked in red (n = 3).

### Antimicrobial Resistance Profiles 2009

The RVLs implemented revised panels and procedures for antimicrobial sensitivity testing from the beginning of 2010. These are in line with international practice, and are designed to support the DAFF's international commitments to report on antimicrobial resistance patterns in food-producing animals whilst also providing valuable information to the practicing veterinary clinician.

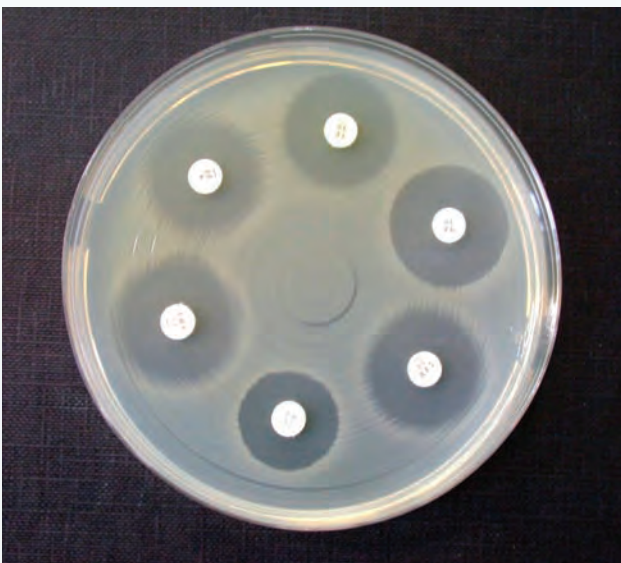


Figure 68: Antimicrobial resistance testing showing zones of clearance (Photo: Kevin Kenny).

### Milk Pathogens

#### *Streptococcus dysgalactiae*

The results of *in-vitro* antimicrobial susceptibility and resistance testing for *Streptococcus dysgalactiae* isolates from bovine milk samples are shown in Figure 69.

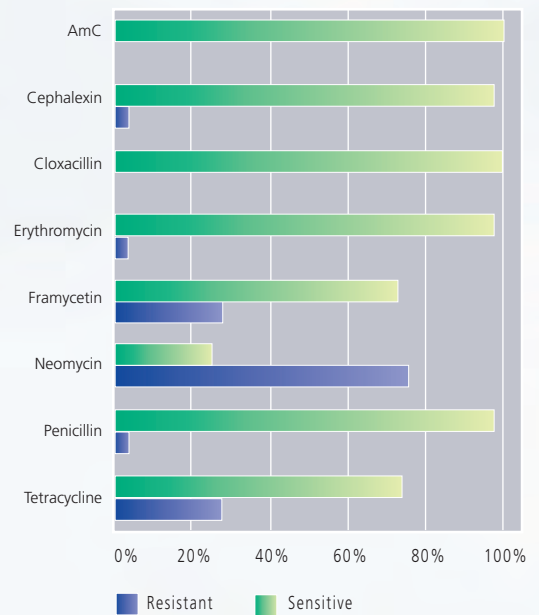


Figure 69: Antimicrobial resistance in *Strep. dysgalactiae* isolates from bovine milk submissions in 2009 (n = 209) (AmC = amoxicillin-clavulanate).

No resistance was recorded to amoxicillin-clavulanate or cloxacillin - with only a few isolates showing resistance to penicillin, cephalixin, or erythromycin. These results are similar to those reported in the RVL Disease Surveillance Reports for 2006 to 2008.

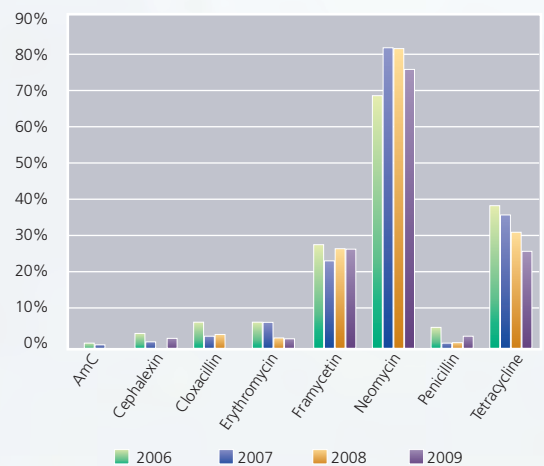


Figure 70: Antimicrobial resistance *in-vitro* in *Strep. dysgalactiae* isolates from bovine milk submissions for the years 2006 to 2009 (n = 700) (AmC = amoxicillin-clavulanate).

Figure 70 illustrates the very similar resistance patterns recorded for *Streptococcus dysgalactiae* isolates over each of the four years 2006 to 2009. Fourteen per cent of isolates were sensitive to all eight antibiotics on test – and 52 per cent were sensitive to all but one. Resistance was most frequently recorded against neomycin (76 per cent), followed by tetracycline (33 per cent).

### Escherichia coli

Antibiotic susceptibility and resistance patterns for *E. coli* isolates from bovine milk samples are shown in Figure 71.

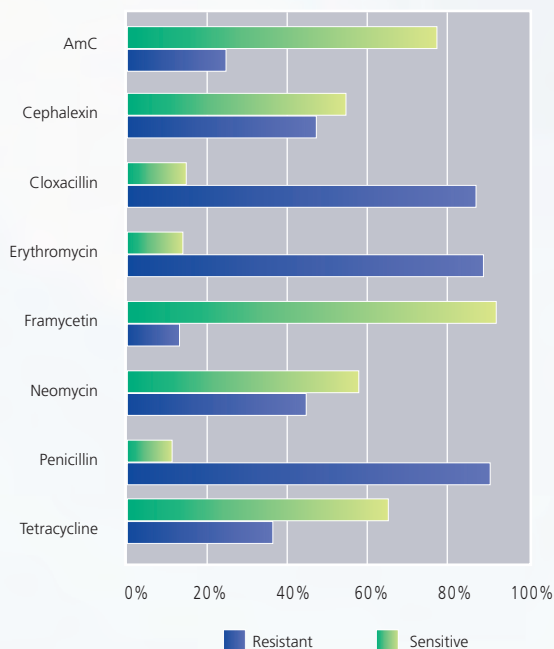


Figure 71: Antimicrobial resistance in *E. coli* isolates from bovine milk samples in 2009 (n = 517) (AmC = amoxycillin-clavulanate).

Isolates showed a relatively high degree of resistance. Over 80 per cent of isolates registered resistance to one or more of the antibiotics cloxacillin, erythromycin and penicillin. Over 40 per cent were resistant to cephalixin and/or neomycin. Framycetin showed the least degree of resistance at 10 per cent.

It should be noted, however, that as many coliform isolates from milk samples are due to sampling error or environmental contamination, antibiotic susceptibility results for *E. coli* must be interpreted in the light of the presenting clinical syndrome.

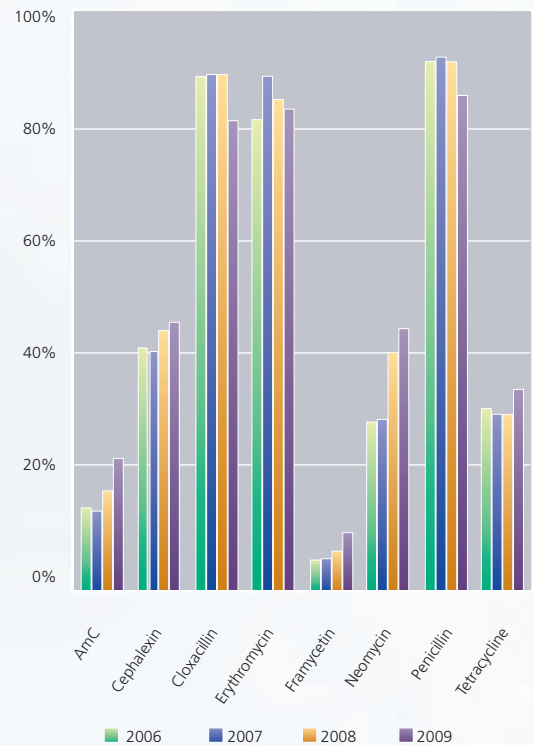


Figure 72: Antimicrobial resistance *in-vitro* in *E. coli* isolates from bovine milk samples for the years 2006 to 2009 (n = 1,385) (AmC = amoxycillin-clavulanate).

While comparison of the results over the four-year period 2006 to 2009 showed little evidence of an increasing trend in resistance by *E. coli* (Figure 72), analysis of the data showed considerable multi-drug resistance in isolates. Sixty per cent of *E. coli* isolates were resistant to four or more of the eight antibiotics on test. On the other hand, over 99 per cent of isolates over the four-year period were sensitive *in-vitro* to at least one of the four antibiotics framycetin, amoxycillin-clavulanate, tetracycline, or neomycin.

## Enteric Pathogens

### Salmonella Dublin

Resistance levels for *Salmonella* Dublin isolates from bovine submissions for each of the four years from 2006 to 2009 are shown in Figure 73. The results show that a fairly wide choice of antibiotics is available for the treatment of *Salmonella* Dublin, based on *in-vitro* testing and that *Salmonella* Dublin isolates typically show only limited antimicrobial resistance.

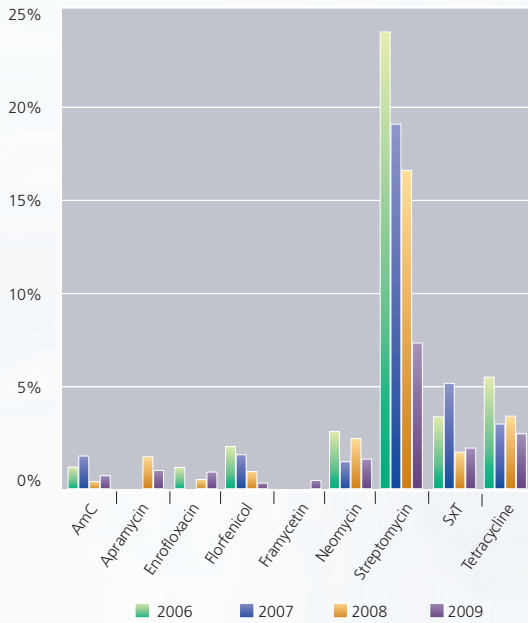


Figure 73: Antimicrobial resistance in *Salmonella* Dublin isolates from bovine submissions for the years 2006 to 2009 (n = 356) (AmC = amoxicillin-clavulanate, SxT = sulphamethoxazole-trimethoprim).

### Salmonella Typhimurium

Antimicrobial susceptibility profiles for *Salmonella* Typhimurium isolates in the Regional Laboratories in 2009 are shown in Figure 74 (multiple animal species). There is a broad degree of sensitivity, with little or no resistance recorded to the antibiotics enrofloxacin and framycetin.

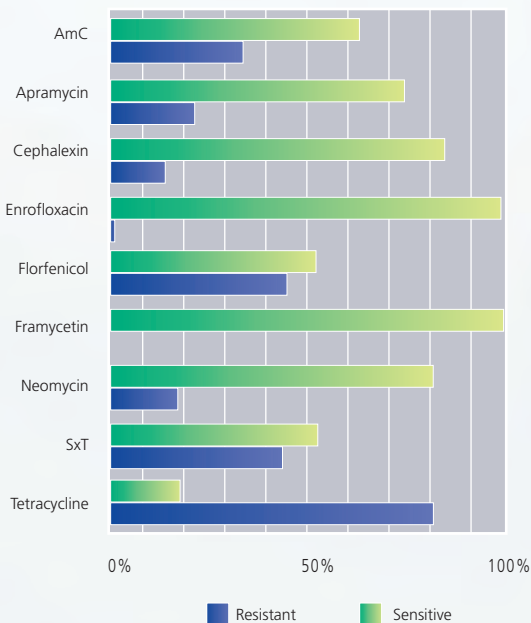


Figure 74: Antimicrobial resistance in *Salmonella* Typhimurium isolates from avian, bovine, ovine, porcine, and equine submissions in 2009 (n = 53) (Sxt = sulphamethoxazole-trimethoprim, AmC = amoxicillin-clavulanate).

Resistance patterns for *Salmonella* Typhimurium isolates from bovine submissions for each of the four years from 2006 to 2009 are shown in Figure 75 (192 isolates). There is no evidence of a pattern in terms of increasing or decreasing resistance in the case of any antibiotic. In absolute terms, resistance was least likely to be encountered to enrofloxacin (187 of 190 isolates sensitive) followed by framycetin (140 of 192 isolates sensitive) and amoxicillin. However, cephalixin, which was tested less frequently, scored second best in percentage terms with 76 per cent of 42 isolates tested showing sensitivity *in-vitro*. The highest level of resistance was to tetracycline (64 per cent), followed by apramycin and sulphamethoxazole-trimethoprim.

The majority of the 36 *Salmonella* Typhimurium isolates phage-typed in 2009 (19 bovine, one porcine) were DT104 or DT 104b as has been the trend in Ireland in recent decades.

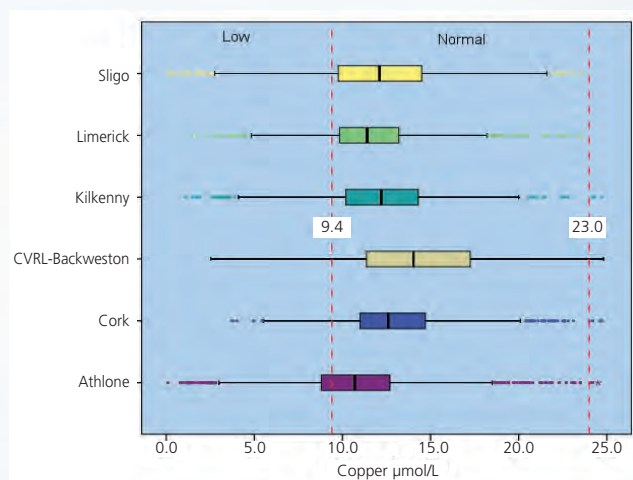


Figure 75: Antimicrobial resistance in *Salmonella* Typhimurium isolates from submissions (multiple animal species) for the years 2006 to 2009 (n = 192) (Sxt = sulphamethoxazole-trimethoprim, AmC = amoxicillin-clavulanate).

## Clinical Chemistry

### Copper and Selenium Analyses in the Regional Veterinary Laboratories in 2009

The Regional Laboratories and the Central Veterinary Laboratory carried out copper analyses on a total of 11,686 bovine blood samples in 2009. A value of 9.4  $\mu\text{mol/L}$  or less indicates low or deficient serum copper levels.



**Figure 76:** A box plot showing the range of bovine serum copper values recorded in the six RVLs in 2009. The boxes represent the range in which 50 per cent of the values lie. The normal range of 9.4 to 23  $\mu\text{mol/L}$  is shown (n = 11,686).

Analysis of the results revealed that 2,167 (18.5 per cent) samples were recorded with a copper level which was deemed to be below the normal range. Athlone RVL recorded the highest number of deficient samples (Figure 76), which is probably a reflection of the molybdenum levels in soils in the midlands. Sligo RVL recorded the second highest number of copper deficient samples.

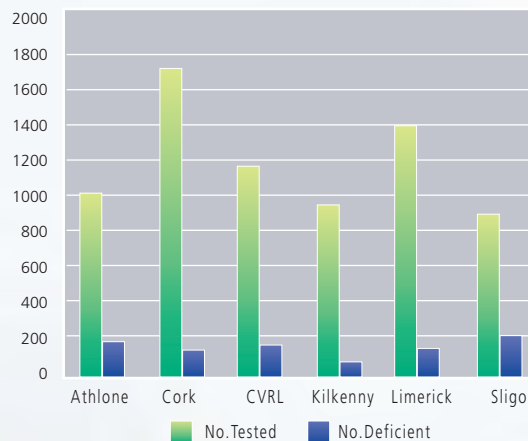


**Figure 77:** Poor thrive and change in hair pigmentation in a two year old heifer associated with copper deficiency (Photo: John Fagan).

Copper deficiency is often the result of an excess of the mineral molybdenum in the soil. Molybdenum binds copper, making it unavailable for absorption from the gastrointestinal tract of the ruminant. As well as affecting thrive, copper deficiency may cause skeletal abnormalities, reproductive difficulties, impaired nervous tissue function, and changes in hair and skin pigmentation (Figure 77).

Selenium is an essential component of a number of proteins, enzymes and hormones in ruminants. In cattle, selenium deficiency is associated with a wide range of health and production problems which include ill-thrift, retained foetal membranes in cows, infertility, and impaired immunity. The selenium status of an animal can be determined by measuring the activity of the selenium-containing enzyme glutathione peroxidase (GSH-Px) in red blood cells, or by directly measuring blood (elemental) selenium levels. There is a good degree of correlation between both methods of measurement. Selenium status is also routinely measured by analysis of kidney selenium concentrations of carcasses presented for *post mortem* examination.

Blood samples for elemental selenium or GSH-Px analysis must be submitted in Lithium Heparin bottles (green-topped).



**Figure 78:** The number of bovine samples analysed for determination of selenium status (either by blood selenium analysis or measurement of GSH-Px activity), and the numbers of those samples identified as deficient in each of the RVLs<sup>12</sup> in 2009 (n=7009).

Figure 78 shows the results of blood selenium and GSH-Px analyses, categorised by laboratory, during 2009. These results reflect the long-term status of selenium in these animals - as both blood GSH-Px and elemental selenium rise and fall slowly with changing tissue selenium levels. The blood selenium status in a herd or region tends to reflect the soil selenium status in that region. Factors such as soil pH, soil sulphur content, and heavy rainfall can all influence the soil selenium status - and may explain some of the variations between the catchment areas served by each RVL.

<sup>12</sup> Samples submitted to Dublin RVL are analysed in the Clinical Chemistry Section, CVRL, Backweston.

A blood selenium value of less than or equal to 0.75  $\mu\text{mol/L}$ , or a GSH-Px level of less than 40 units/ml PCV, is indicative of low or deficient selenium status. Similar to the pattern in 2008, Sligo RVL recorded the highest proportion of selenium deficient samples (23.9 per cent), while Cork RVL recorded the lowest proportion of selenium deficient samples (9.1 per cent), in 2009.

## Haematology Testing in the Regional Veterinary Laboratories

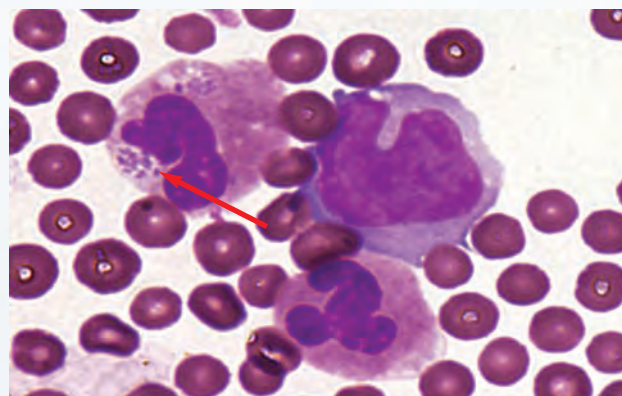
Haematology testing is available in all the Regional Veterinary Laboratories. It is a very useful tool for the clinician in assessing the adequacy of haematopoiesis, or the presence or absence of a systemic inflammatory response in the sampled animal. An EDTA blood sample is required by the laboratory to perform a full differential haematological examination. This should reach the laboratory within 24 hours of collection – preferably within 12 hours - to allow the full range of red and white cell analyses to be performed. The fresher the sample, the more reliable are the results – in particular in relation to white cell parameters, as white cells are more fragile than red cells, and degrade rapidly after collection.

Examination of blood films can also confirm the diagnosis of haemoparasitic diseases. Presented below is a description of a case of tick-borne fever in a dairy herd - where haematological examination was central to achieving a diagnosis.

### Tick-borne Fever in a dairy herd

Limerick RVL investigated an animal health problem on a 55-cow dairy farm in late April 2009. The farm was fragmented. The grazing pasture consisted of approximately 80 acres, twenty acres of which were prone to flooding. Affected cows were mostly first or second calvers. They presented clinically with high temperatures, milk drop and depression. A clear nasal discharge was seen in a small number of affected cows. New cases occurred every few days, and were easily recognised by the farmer. The animals responded well to antibiotic therapy, but the milk yield was slow to recover to normal levels.

Examination of the farm records (which were meticulously kept) showed that a similar syndrome had occurred on the farm every year at roughly the same time, i.e. a few weeks after turnout. Different respiratory virus vaccination protocols had been implemented, but did not seem to have any effect on preventing recurrence of the syndrome.



**Figure 79:** A blood film from a cow with tick-borne fever showing a neutrophil with morulae (arrow) of *Anaplasma phagocytophila* in the cytoplasm (Photo: Jim O Donovan).

Blood samples (in EDTA) were taken from a number of the recently affected cows and routine haematology testing was carried out. Smears of each sample were also made on glass slides, stained and examined microscopically. Intracellular parasitic inclusions were seen in many of the white blood cells (mostly neutrophils) examined (Figure 79). The appearance of these inclusions was consistent with *Anaplasma phagocytophila*, a rickettsial organism carried by the tick *Ixodes ricinus*. A diagnosis of tick-borne fever was made.

Tick-borne fever (TBF) tends to be seasonal in occurrence, linked with the feeding activity of the tick vector. The organism can survive in infected ticks for long periods of time.

Following infection, the incubation period is typically between six and fourteen days. Neutrophilia develops two days post-infection and is followed by severe leukopenia due to lymphocytopenia and neutropenia, thus increasing susceptibility to secondary infections.

Effective control can be achieved by eliminating or markedly reducing contact with the tick vector, either by grazing cattle on tick-free pastures in lowland areas, or by the use of treatments effective against *Ixodes ricinus*. There are no effective vaccines available to protect ruminants from clinical TBF.

## Proficiency Testing in the Regional Veterinary Laboratory Service

The five Regional Veterinary Laboratories and Clinical Chemistry Section, Backweston (for Dublin RVL), subscribe to four Proficiency Testing (PT) Schemes. Three schemes are operated by the Veterinary Laboratory Agency, UK (haematology, microbiology and tissue lead and copper). The Randox International Quality Assessment Scheme (RIQAS) offers biochemistry proficiency testing for protein, blood metabolites, enzymes, and major and trace element tests.



Proficiency testing for bacteriology involves a freeze-dried material containing a known pathogen being sent to all participating laboratories with a limited case history. Each laboratory is asked to identify the pathogen and is then scored on the basis of its results.

Proficiency testing for the haematology and biochemistry components of RVL work involves each laboratory testing sample materials for certain specified constituents (red and white cell parameters, blood metals, etc.). The returned results for all of the laboratories in the scheme are assessed by the external proficiency supplier (i.e. VLA or RIQAS). After obvious 'outlier' values have been discarded, a consensus mean is arrived at. Each laboratory then receives its own individual results - together with a statistical analysis showing how its performance compares to the mean for the peer group. This process allows any laboratory with a result of two or more standard deviations from the consensus mean for any one component to investigate its analytical procedures. Participation by RVLs in PT schemes is beneficial in excluding the possibility that laboratory results could be biased in a particular direction - and is one of the requisites for accreditation by the Irish National Accreditation Board (INAB). All of the Regional Laboratories also follow an internal quality control programme using standard reagents and controls.

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

Compliance with correct procedures for the packaging of samples being submitted to the Laboratory Service is vital in 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:

<http://www.unecwe.org/trans/danger/publi/adr/adr2007/07C0ontentsE.html>.



Figure 80: Wrap the sample in absorbent material and place in a leak proof plastic container.

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 (Figure 80).

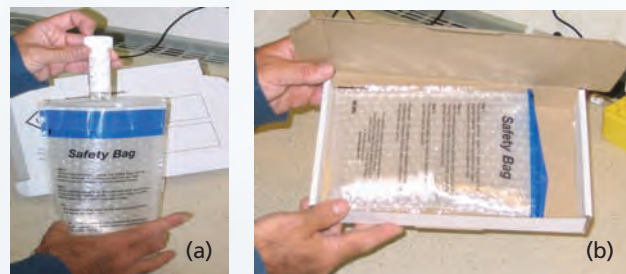


Figure 81 (a) & (b): Place the leak proof plastic container in an outer padded envelope.

This is then placed in the outer padded envelope and sealed (Figure 81).

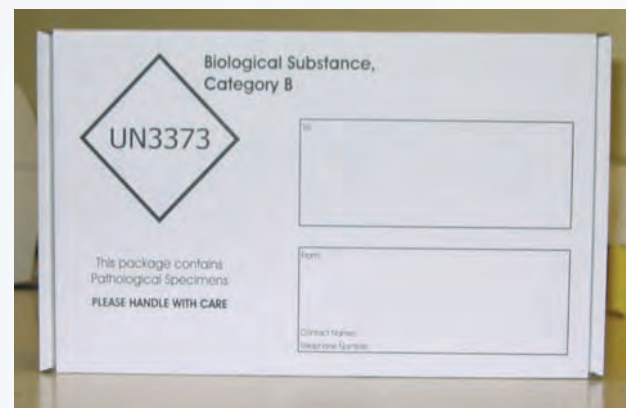


Figure 82: The words "Diagnostic Specimen" or "Pathological Specimen" must be labelled on the outside of the package.

The words "Diagnostic Specimen" or "Pathological Specimen" must be labelled on the outside of the package (Figure 82).

Contact details for suppliers of appropriate packaging materials may be obtained from the Institute of Packaging Ireland (also known as the Irish Packaging Society).

## Class A Disease Surveillance 2009

### Foot-and-Mouth Disease

There have been no cases of Foot-and-Mouth (FMD) disease in Ireland since 2001. FMD is a highly contagious viral disease of cloven-hooved animals characterised by the typical clinical findings of high fever and the formation of vesicles inside the mouth and on the feet. The vesicles may burst leaving ulcers (Figure 83).

Consequent drooling and lameness are common signs. The virus has an incubation period of between one and 12 days. While mortality among infected animals is low, morbidity is very high with significant production loss and impediments to international trade. FMD is a statutorily notifiable disease. There were no FMD suspects notified to the Department during 2009.



**Figure 83:** A Foot-and-Mouth lesion with sloughing of the mucosa and fibrin deposition in an adult cow (Photo: Ger Murray).

### EuFMD Training in Erzurum, Turkey

The Food and Agriculture organisation (FAO), supported by the EU (DGSANCO), have provided training to veterinarians from European Commission for the Control of FMD (EuFMD) states. The aim of this training, in which some RVL veterinary staff have participated, is to provide state veterinarians with experience in the clinical recognition of FMD in ruminants, as well as in the processes of investigation and decision making in the face of an outbreak. The participation of Department of Agriculture veterinary staff facilitates the maintenance of a group within the organisation who have up-to-date experience of FMD investigation and epidemiology.



**Figure 84:** (a) and (b) show the use of a probang to obtain a pharyngeal fluid sample for FMD virus detection. (c) shows the pen-side test used for on-farm testing of samples for FMD virus (Photos: Mícheál Casey).

The course comprises a week of field-based investigations near Erzurum in eastern Anatolia, Turkey. FMD is endemic in some regions of eastern Turkey, and vaccination is practiced twice yearly as a means of control in cattle - with approximately 85 per cent coverage of the population. There are seven FMDV serotypes: O, A, C, SAT-1, SAT-2, SAT-3, and Asia-1 of which serotypes O and A are endemic in Turkey. There is no cross-protection between serotypes and even within serotypes there is considerable variation between strains. Because of this, vaccines must be highly specific to the strain which is endemic in an area to be effective.

The husbandry systems in the mountainous areas of eastern Anatolia differ considerably to those in Ireland. Most notably, communal grazing is practiced which facilitates rapid spread of disease when an outbreak occurs. While herd sizes are smaller, this grazing system requires that the village rather than the herd be considered the basic epidemiological unit for the purposes of disease control.

### Bluetongue

Bluetongue is a viral disease of ruminants. There are 24 known serotypes of the causative virus and it is spread between animals - primarily by midges. The midge *Culicoides imicola* is the primary vector for the virus, but the virus has also adapted to other midge species. In Europe the disease was formerly restricted to the Mediterranean basin but in recent years several serotypes have spread into many countries in Northern Europe. Apart from the midge vector, other lesser means of spread of the Bluetongue virus such as transplacental spread, spread by infected semen, or iatrogenic spread (e.g. syringe needles), may also play a role in the transmission of the virus between animals. Bluetongue Serotype 8 was associated with outbreaks in many European countries, including the UK. Incubation of the virus takes between four and 20 days after which a clinical disease of variable severity and mortality may occur. 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 at <http://www.agriculture.gov.ie/bluetongue>.

Surveillance for Bluetongue is ongoing. Bluetongue testing was carried out on 3,956 samples in the Virology Division, Central Veterinary Research Laboratory (CVRL) Backweston during 2009. These samples represent all susceptible animals imported from areas where the disease is present, and a random sample of susceptible Irish animals. Ireland continues to be Bluetongue-free.

## Avian Influenza

There is an active program of surveillance to prevent the introduction of highly pathogenic avian influenza (HPAI) into Ireland. HPAI is a highly infectious viral disease which affects the respiratory, digestive or nervous systems of many species of birds. As well as posing serious risks to avian health, the disease can have potentially serious consequences for human health. Carcass submissions from HPAI suspects are initially examined in the RVLs by laboratory staff. Samples are then selected by RVL pathologists for analysis by Virology Division, CVRL Backweston. During 2009, 444 samples from birds were tested in CVRL for HPAI by real-time PCR. All of these were negative for the presence of HPAI. In addition to these, samples were also tested from another 177 wild birds. Two of these specimens were positive for the Matrix gene of HPAI virus, one of which (from a wigeon in Co. Louth) was characterised as subtype H5, while the second specimen (from a teal in Co. Louth (Figure 85)), could not be further characterised.

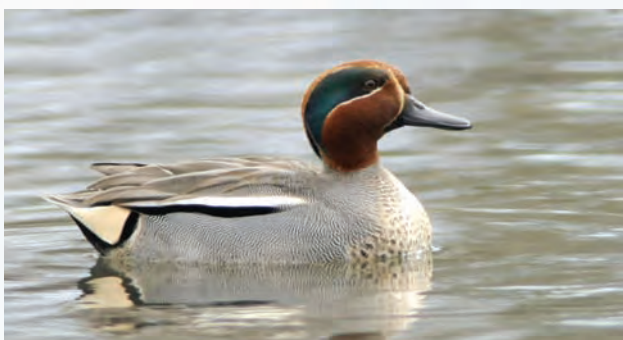


Figure 85: A teal (*Anas crecca*) (Photo: Michael Finn/Birdwatch Ireland).

Two hundred and eighty one of the 444 samples which were tested by real-time PCR were further submitted for HPAI virus isolation, all of which were negative. The introduction of HPAI remains a significant threat to the poultry industry.

## Bovine Spongiform Encephalopathy (BSE)

In 2009, confirmatory diagnosis was carried out in the TSE National Reference Laboratory (NRL) in the Central Veterinary Research Laboratory (CVRL) on samples from a total of 53 bovines. Nine samples were confirmed as positive in the NRL – all of these had been initially detected in the private rapid testing laboratories as part of the DAFF active surveillance program.

The remaining 44 samples examined in the NRL were clinical suspect cases submitted via the RVLs (passive surveillance). The brains were removed from these animals in the RVLs and submitted to the NRL for confirmatory diagnosis. All of these were confirmed as negative for BSE in the NRL. Table 10 shows a breakdown of the histopathological diagnoses reached for each of these BSE-negative cases.

Histopathological Diagnosis	Number
No specific lesion	22
Listerial encephalitis	15
Non-suppurative encephalitis	3
Neoplasia	2
Fibrinosuppurative ventriculitis	1
Progressive ataxia of Charolais cattle	1
Total	44

Table 10: The histopathological features of 44 adult bovine brains from clinical BSE suspects examined in 2009.

The nine positive cases detected in 2009 compare favourably with the figures for 2008 when 23 cases were confirmed as positive, and reflect the continued decreasing trend in positive cases witnessed since the peak in 2002.

## Scrapie

Scrapie is a fatal transmissible disease of sheep and goats and, like BSE in cattle, belongs to the group of conditions referred to as Transmissible Spongiform Encephalopathies (TSEs).

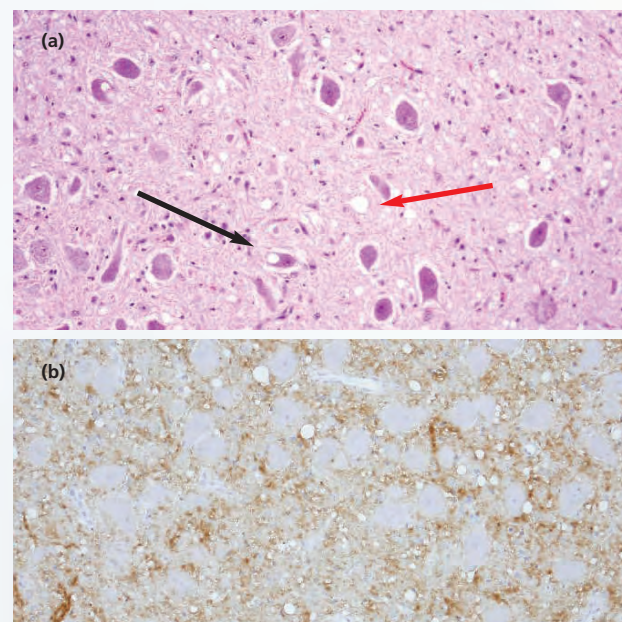


Figure 86: (a) Vacuolated neuron (black arrow) and vacuolation of the neuropil (red arrow) in the dorsal vagal nucleus of a sheep with classical scrapie (H&E, 100x). (b) Positive immunostaining (brown staining) in the dorsal vagal nucleus of the same sheep (100x) (Photos: Máire McElroy).

Clinical signs of Scrapie typically include weight loss, altered gait or posture, and skin irritation resulting in pruritus. It is naturally transmissible between sheep and goats, and clinical signs are normally observed in sheep at three to four years of age - although they have been recorded occasionally in younger sheep.

The Department of Agriculture, Fisheries and Food operates the Scrapie Monitored Flock Scheme which is voluntary and is targeted at pedigree flocks. This scheme is designed to provide for DAFF regulation of holdings seeking to fulfil specific requirements for the trading of breeding sheep and goats within the EU as laid down in Annex VIII of Regulation (EC) No. 999/2001. The Department also has an active Scrapie surveillance programme which involves the testing of samples collected from sheep at slaughtering plants and knackeries.

The most common type of Scrapie in Ireland is known as Classical Scrapie. New Scrapie strains, distinct from Classical Scrapie, have been identified since 1998, and are referred to as Atypical Scrapie. Atypical Scrapie does not appear to be as transmissible as Classical Scrapie.

In 2009, 38 cases of Scrapie were confirmed by the NRL. Of these, 33 were classified as Classical Scrapie (Figure 86), while the remaining five were classified as Atypical Scrapie (Nor-98).

The 33 confirmed cases of Classical Scrapie came from eight separate flocks; 23 were submitted for confirmatory testing from a rapid testing laboratory (active surveillance), while the remaining 10 were submitted as clinical suspect cases (passive surveillance) through the RVLs. Two further clinical suspect cases submitted through the RVLs were confirmed as being negative for Scrapie.

Of the Atypical Scrapie cases, tissues from four out of the five were sent to the NRL from rapid testing laboratories i.e. they were detected through the active surveillance programmes. Tissues from the remaining atypical case were sent to the TSE NRL from an RVL under the Scrapie Monitored Flock Scheme.

During 2009, twelve new flocks were identified with scrapie; seven with Classical Scrapie and five with Atypical Scrapie (Nor-98). Co-existence of the classical and atypical form of the disease was not detected in any flock.

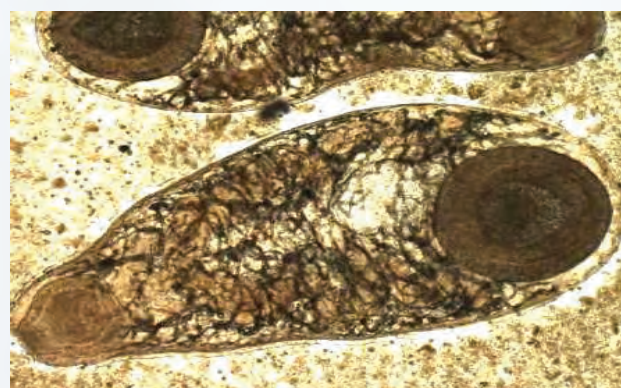
All samples tested by Discriminatory Western Blotting (to differentiate between scrapie in sheep and BSE in sheep) were reported as 'Scrapie-like' in 2009 - thus providing evidence for the continuing absence of BSE in sheep in Ireland.

## A Selection of RVL Farm Investigations in 2009

An important role of RVL pathologists is the provision of advice regarding animal disease and production problems. Laboratory pathologists carry out on-farm investigations in cases where zoonotic, exotic or novel diseases are suspected to be involved. Investigations may also be undertaken where production or infectious endemic diseases have given rise to significant on-farm losses. Presented below are summaries of selected farm investigations conducted by RVL veterinary staff during 2009.

### Mortality in a sheep flock due to Paramphistomosis

In November 2009, Athlone RVL conducted a farm investigation on a sheep flock of 47 ewes which were grazing an out-farm for the previous two weeks. The first death in the group was a day after arrival at the out-farm, and a further 14 died over the following two weeks - most being recorded as sudden deaths. The out-farm had been grazed by ewes and lambs during the summer without any signs of illness. Flukicide had been administered on a regular basis to the flock, and multivalent clostridial vaccine had also been administered. Virtually all of the flock, with the exception of only a few, were scouring on the day of the visit. There was evidence of mild to moderate dehydration in many of the animals examined. Eight animals were selected at random and sampled. Notable laboratory results included marked hypoalbuminaemia in all eight animals. Examination of a faecal smear identified *Paramphistomum* spp. larvae (rumen fluke) in the faeces in large numbers (Figure 87).



**Figure 87: *Paramphistomum* spp. larvae identified in a faecal smear from a ewe (Photo: Jim O Donovan).**

Paramphistome adults tend to cause mild illthrift - but the larvae can cause significant upper small intestinal enteritis with marked hypoalbuminaemia. Morbidity can reach 100 per cent and mortality of 25 per cent is reported. The life cycle is similar to that of liver fluke. In heavy infections, development of the young flukes

may be retarded, and they can stay in the small intestine without progressing to the rumen for more than four months, causing severe enteritis. Diagnosis is dependant on finding eggs in the faeces (larvae are rarely found in faeces), as well as the response to treatment (oxyclosanide or radoxinide). Following dosing of these animals with oxyclosanide, all of the flock had ceased scouring within three days and no further cases occurred.

### Immunosuppression in a suckler herd

Sligo RVL was asked to investigate an outbreak of enteritis and respiratory disease in calves in a herd where vaccination for enteric disease, BVD, and respiratory disease was practiced. Coronavirus and rotavirus had been detected in faecal samples submitted to the laboratory. There had also been an outbreak of respiratory disease the previous December, and morbidity on farm was high for both respiratory disease and enteritis. BVD virus was identified in one of two calves submitted for *post mortem* examination. A weanling heifer purchased the previous autumn was also found to be persistently infected (PI) with BVD virus. Interestingly, this heifer was in good body condition and showed no evidence of ill thrift. Following confirmation of her persistently infected status she was slaughtered.

In the course of the visit the ventilation of the animal housing was assessed. While a number of deficiencies were identified, ventilation was not considered to be a significant contributory problem. As the increase in morbidity on farm had coincided with the introduction of the PI animal and the herd health situation improved following its removal, there would appear to be an association between the disease problems encountered in this herd and the BVD virus infection. This case clearly shows the potential consequences of introducing persistently infected animals into a herd, as well as the role of BVD in the occurrence of other disease problems on farms. It also shows that BVD vaccination alone is not sufficient to control the disease.

### Actinobacillosis in a herd of bullocks

Athlone RVL was asked to conduct an investigation in a herd in which a quarter of the herd exhibited abscessation of varying severity in the region of the submandibular lymph node. A small number had also developed abscesses on the shoulder. The enterprise comprised 126 bullocks which were kept in slatted housing for most of the year. The problem was first noted in 2007. However, in 2008 the numbers affected had increased considerably. The animals affected in 2008 did not have any direct contact with those affected in 2007. All animals had been slaughtered in the autumn, with a six week interval when no animals were kept on the farm. Some of the bullocks which had

not shown abscessation were slaughtered and had *Actinobacillus* spp. lesions in the lymph nodes. Hyper-salivation was not a feature of the disease. Blood copper and selenium levels were very low in all animals sampled during the visit. There appeared to be two substantial issues – firstly the question of herd immunity and the ease with which the infection seemed to have passed through the herd, and secondly, the need to break the cycle of infection. *Actinobacillus* sp. was isolated from some of the lesions, confirming a diagnosis of Actinobacillosis. It was concluded that the low copper and selenium status was compounding the problem by reducing the immunity of the herd to infection. The herdowner was advised of the importance of segregating infected cattle. Advice was also given regarding adequate disinfection of the sheds after the cattle were let out - using a power washer and appropriate disinfectant - with particular attention being given to areas where cattle might scratch themselves. The shoulder abscesses in the bullocks examined did not appear to be associated with the adjacent (pre-scapular) lymph node, and were assumed to be acquired from scratching against the walls of the shed. Advice was also given regarding copper and selenium supplementation.

### Psoroptic mange in a Belgian Blue bull

*Psoroptes ovis* (previously known as *Psoroptes bovis*) was found in a skin scraping submitted to Sligo RVL in February 2009 (Figure 88). A farm visit was conducted to investigate the case further. The affected animal was a 23 month old pedigree Belgian Blue bull, which had been purchased in a dispersal sale the previous August. The Belgian Blue breed seems to be particularly susceptible to this condition. There was severe pruritus associated with dermatitis on the dorsum, the hindquarters, and the medial aspect of the hocks. The hair loss was quite pronounced (Figure 89). The owner also reported that the affected animal had lost weight.

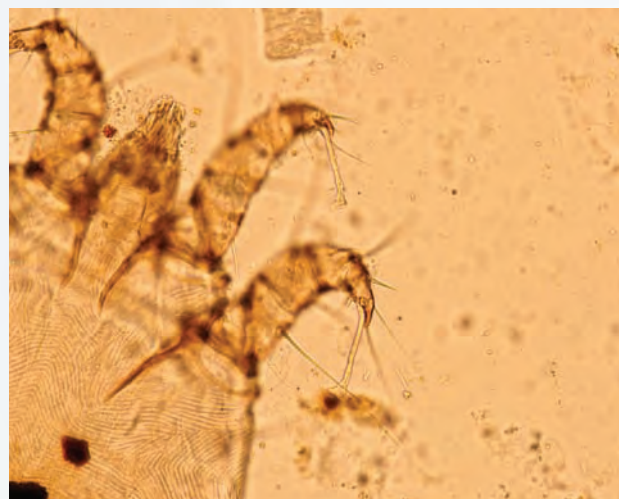


Figure 88: *Psoroptes ovis* identified on a skin scraping from a 23 month old pedigree Belgian Blue bull (Photo: Damien Barrett).

A group of Belgian Blue cross weanlings that shared a pen with the affected bull were examined. Two of the animals showed signs of hair loss. Skin scrapings were taken from both animals, but *Psoroptes* spp. were not detected.



**Figure 89: Alopecia in the perineal area of a Belgian Blue bull due to *Psoroptes ovis* infection (Photo: Damien Barrett).**

Sheep were also kept on the premises, but there was no evidence of pruritus in the sheep. Strains of *P.ovis* that infect cattle and sheep are species specific, so cross infection would not be expected.

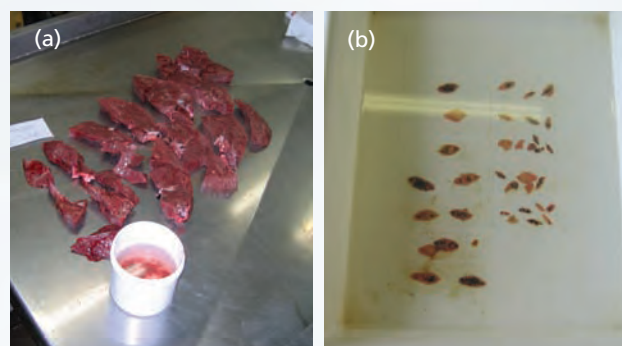
The recommended treatment for the condition is three doses of ivermectin at 200µg/ kg at 14 day intervals. A single dose of ivermectin is insufficient, as it suppresses rather than eliminates *Psoroptes* spp.

Subsequent to this investigation, there were a number of reports across the country of Psoroptic mange in cattle which had originated in the same herd as the bull affected in this case. All were treated successfully.

### An investigation into suspected triclabendazole-resistance in a sheep flock with chronic liver fluke disease (*Fasciola hepatica*)<sup>13</sup>.

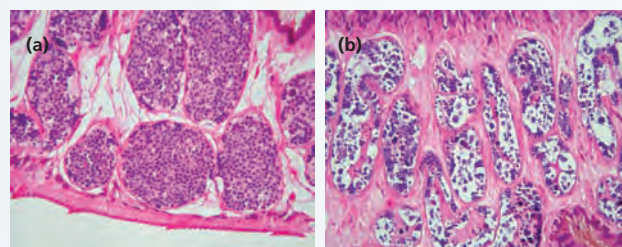
The background to this investigation was high mortality in a flock of adult ewes caused by severe acute fasciolosis - and occurring in both early and late 2009. This was despite repeated dosing with triclabendazole (TCBZ). Conventional parasitological methods to demonstrate resistance to anthelmintics such as the faecal egg reduction test are not sufficient to demonstrate TCBZ resistance by liver fluke. The classic approach is a complicated, multi-step

procedure as it requires egg-hatch and miracidial mobility tests, as well as testing for the ability of larvae to develop in snails and produce infective metacercariae. An alternative approach, which has been used in an experimental setting by Dr. Bob Hanna and colleagues at Agri-food and Biosciences Institute (AFBI), Stormont, is to assess morphological features of the reproductive organs of TCBZ-treated flukes. Susceptible flukes exhibit degenerative features in these organs at specific time intervals after treatment with TCBZ, whereas no such changes are observed in TCBZ-resistant flukes when treated with the same compound.



**Figure 90: (a) Liver of a fluke-infested sheep killed 3 days after treatment with triclabendazole and dissected to recover flukes. (b) Flukes recovered from the liver are flat-fixed in formaldehyde (Photos: Dónal Sammin)**

Using this method to demonstrate TCBZ resistance in the field involved the following steps: (1) treatment of two fluke-infested sheep with TCBZ (the sheep were weighed and dosed according to the manufacturer's recommendations); (2) euthanasia of one sheep at 3 days and one sheep at 3 weeks after treatment; (3) harvesting of flukes from the liver and bile ducts immediately after euthanasia of the host animal and fixation by immersion in formaldehyde (Figure 90 (a) and (b)); (4) examination of the reproductive organs of flukes for the presence of morphological changes induced by TCBZ (by light microscopy) – the absence of such changes indicating resistance to TCBZ (Figure 91).



**Figure 91: Photomicrograph of testes from (a) an untreated liver fluke and (b) from a TCBZ-sensitive liver fluke, 2 days after treatment with TCBZ; extensive drug-induced histopathological changes are evident (Photos: Dr. Bob Hanna, AFBI, Stormont).**

None of the flukes from either sheep showed evidence of TCBZ effects in any of their reproductive organs. The testis profiles were entirely normal, with lots of mature sperm present. The uteri were full of normally-shelled eggs, and also had sperm present amongst the eggs, suggesting recent copulation. Furthermore, most flukes

<sup>13</sup> This investigation was undertaken at Kilkenny RVL with assistance from Dr. Bob Hanna and colleagues at the Agri-food and Biosciences Institute (AFBI), Stormont.

had fresh blood in the gut caecae, showing that they had been actively feeding up until the time of collection. These findings suggests that the fluke population examined were uniformly resistant to TCBZ.

## An Infertility Problem in a Dairy Herd

In April, Athlone RVL investigated an infertility problem on a dairy and suckler enterprise comprising 70 Spring-calving milking cows (averaging approx 1,900 gallons per year) and 45 suckler cows. Eleven of the dairy cows were not pregnant when scanned in the autumn, and the intercalving interval had become wider over the previous two years (with some cows calving in May and June). The infertility problem appeared to be confined to the dairy herd. The herdowner reported several cases of milk fever annually which responded poorly to treatment. Vaccination was undertaken for BVD and Leptospirosis, and supplementation with magnesium and selenium was practiced. Silage was of good quality and maize and meal were fed to the cows all summer. Milk butterfat was recorded in the region of 4 *per cent*.

Test results identified significant antibody titres to *Leptospira* spp. and *Neospora* spp., and serum calcium was low in all animals sampled. All other trace elements were within their normal ranges. Biochemistry results showed that protein balance and energy balance were normal. Culling of the *Neospora*-positive animals was recommended; however it was considered unlikely that Neosporosis alone was responsible for the infertility problem experienced. The calcium result was indicative of a sub-clinical hypocalcaemia problem in the herd. Sub-clinical hypocalcaemia has been associated with decreased immunity in the periparturient cow, as well as a three times greater likelihood of retained foetal membranes. The effect on fertility is also well established, with smaller follicle sizes and longer calving to first service intervals being reported. Potassium levels were also measured and most of the samples had raised values. As the samples showed minimal haemolysis, this result was considered to be indicative of high potassium in the diet. Following further inquiry, the herdowner reported the application of high potash to pastures in the previous year. Potassium levels in the diet can have a significant effect on the absorption of both magnesium and calcium. The DCAB (dietary cation anion balance) method of prevention of milk fever and subclinical hypocalcaemia, which has gained some popularity in recent years, focuses on the potassium level of the diet. At a basic level, this involves the addition of anionic salts to the diet leading to mild acidification which results in better calcium absorption. The herdowner was recommended to review his fertiliser application protocols with a view to reducing potassium intake.

## A Selection of Abstracts from Published Papers 2009

### Nephrotic syndrome due to glomerulopathy in an Irish dairy cow

The *Veterinary Record* 164:179-180 (2009) (Reproduced with their permission).

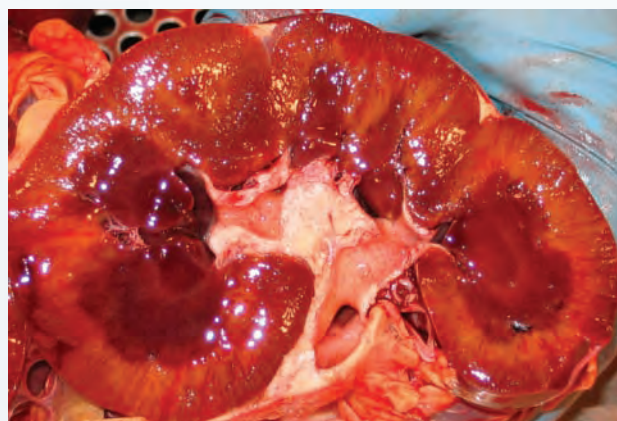
G. M. Murray<sup>1</sup>, and A. E. Sharpe<sup>2</sup>

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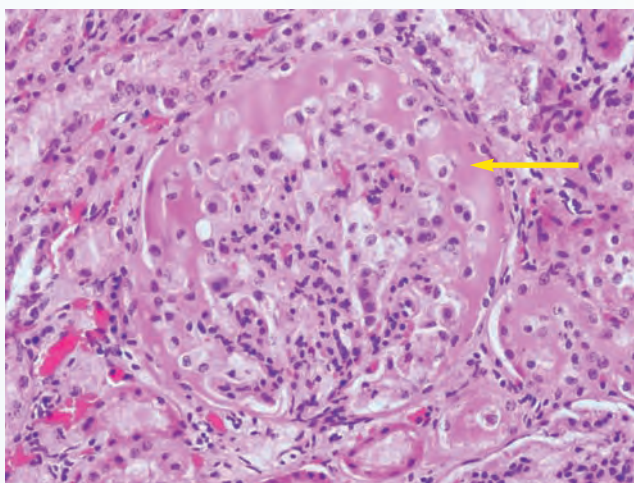
Nephrotic syndrome, a clinical entity characterised by proteinuria, hypoalbuminaemia, oedema and hypercholesterolaemia, results from increased glomerular permeability due to glomerular injury. Although poorly understood, the most commonly identified pathogenesis of this glomerular disease in domestic animals is immune complex deposition where circulating non-glomerular antigen-antibody complexes localise in glomeruli and are visible by electron microscopy.

A three year old Holstein Friesian cow presented with acute diarrhoea, submandibular oedema of two or three days duration and pale mucous membranes. The animal was non-pyrexial and had calved three months previously. The cow was housed, milking well and had no previous history of disease. A tentative diagnosis of chronic fasciolosis was made and a flukicide was administered. There was no response to therapy and the cow died six days later.



**Figure 92: Enlarged bovine kidney with yellow discoloration along the corticomedullary junction (Photo: Ger Murray).**

The *post mortem* examination revealed marked subcutaneous intermandibular oedema spreading to the brisket and low-grade ascites. Both kidneys were mildly enlarged with linear yellow discoloration along the corticomedullary junction (Figure 92). Significant histopathological changes were confined to the kidneys.



**Figure 93: Eosinophilic urinary filtrate containing macrophages in the glomerular urinary space (arrow) (Photo: Ann Sharpe).**

Many glomerular urinary spaces and cortical and medullary tubules were dilated by eosinophilic urinary filtrate (Figure 93). Transmission electron microscopic examination of fixed kidney sections revealed marked ultra structural thickening of the glomerular basement membrane by mainly subendothelial, electron dense deposits. This is the first description of bovine nephrotic syndrome due to glomerulopathy in Ireland. The authors conclude that bovine glomerulopathy may be characterised by minimal histopathological change in glomeruli and should be considered as a differential diagnosis for nephrotic syndrome in cattle.

## The ovine placenta and placentitis - A review

*Veterinary Microbiology* 135 (2009) 90–97 (Reproduced with their permission)

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An appreciation of the complexities of placental structure and function is essential to understanding the pathogenesis of infectious placentitis and abortion. This review aims to illustrate aspects of ovine pregnancy and placentation that will assist both the research worker and the diagnostic pathologist. Morphologically, the ovine placenta is classified as being chorioallantoic, villous, cotyledonary and synepitheliochorial. Apposition of foetal and maternal tissues in early pregnancy eventually leads to the formation of the definitive placenta.

Physiological features of placentation that are essential to normal pregnancy and foetal development include modulation of immune responses at the placental interface, increasing placental bloodflow to allow for increasing foetal demand and the secretion of hormones for the recognition and maintenance of pregnancy. Descriptions of the morphology of the near-term placenta in a normal pregnancy and of the foetal membranes that are voided during normal parturition provide the proper context for understanding the morphological changes associated with placentitis and how these changes are likely to affect placental function.

## Avian influenza viruses detected by surveillance of waterfowl in Ireland during 2003-2007.

*Epidemiology and Infection* (2009) 137: 464-472 (Reproduced with their permission).

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Specimens for the detection of avian influenza virus (AIV) were collected from 1937 waterfowl on the Wexford Sloblands, a major wetland reserve in southeast Ireland, between January 2003 and September 2007. During the same period, 1404 waterfowl were sampled at other locations in Ireland. Specimens were tested either by virus isolation or real-time reverse transcriptase polymerase chain reaction (rtRT-PCR). A total of 32 isolates of AIV, comprising nine subtypes, was obtained from specimens from the Sloblands compared with just one isolate from elsewhere in Ireland. Samples from nine other waterfowl, five of which were from the Sloblands, tested positive for AIV by rtRT-PCR. Ecological factors are likely to have contributed to the higher detection rate of AIV at the Sloblands compared with the rest of Ireland. It was concluded that targeted surveillance at such sites is a cost-effective means of monitoring the circulation of new AIVs in waterfowl, whereas widespread opportunistic sampling is unproductive and wasteful of resources.



## Commensal rodents at farm yards in Co. Cork

*Irish Naturalists' Journal* (2009) 30 (1): 25-26 (Reproduced with their permission).

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In autumn 2007 there were reports of large numbers of 'black' rats on a farmyard in north Co. Cork and also on a neighbouring farmyard. One was a dairy farm while the other was a grain growing enterprise. A total of 520 trap nights were employed trapping rats on both farms which yielded 33 brown rats (11 from the dairy farm and 22 from the grain farm). Using Leslie plots the rat population of each farm was estimated as 16 on the dairy farm and 36 on the grain farm. The dairy farm had a history of *Salmonella* spp. in the herd. On this basis the trapped rats were tested by Cork RVL for *Salmonella* spp. but were all negative.

There were some interesting aspects to these infestations. Firstly there were no black rats found – the last recorded sighting of these animals in Ireland was on Cork docks in 1976. Secondly there was evidence of significant numbers of mice on the farm which goes against the popular belief that rats and mice will not occur together. Finally, other surveys of farm yards in Cork have identified rats on 37 *per cent* of farm yards showing a high incidence of rats on farmyards overall.

## Demographics of cattle positive for *Mycobacterium avium* subspecies *paratuberculosis* by faecal culture, from submissions to the Cork Regional Veterinary Laboratory

*Irish Veterinary Journal*, Volume 62 No. 6:398-405. (Reproduced with their permission)

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The demography of bovine infections caused by *Mycobacterium avium* subspecies *paratuberculosis* (MAP) in Ireland is poorly defined. The objective of this study was to describe the demographics of cattle positive to MAP on faecal culture, based on submissions to the Cork Regional Veterinary Laboratory (Cork RVL) from 1994 to 2006. The study focused on all available faecal samples from adult cattle with non-responsive chronic diarrhoea that were submitted by private veterinary practitioners to Cork RVL for MAP culture. For each MAP-positive by faecal culture animal, data were collated from Cork RVL and Cattle Movement Monitoring Scheme (CMMS) records. Johne's disease (JD) was confirmed in 110 animals from 86 herds by the Cork RVL between 1994 and 2006, with a rate of positive cases between 15 *per cent* and 18 *per cent* over the last four years of the study. Two breeds (Holstein/Friesian or Limousin) made up 78 *per cent* of submissions. Movements were assessed for the 57 study animals with available movement information, 90 *per cent* died within one year of the test and 26 *per cent* tested positive in the herd they were born into. The study provides preliminary information about movement trends and demographics of animals with MAP positive submissions. Although the study area is restricted, it includes the most intensive (and economically-important) dairy region in Ireland. The demographics of JD infection from the study area are in agreement with international reports. Further work is required to determine demographic trends, incidence and prevalence of JD throughout Ireland. It is hoped this work may contribute to the development of a surveillance strategy for MAP by Regional Veterinary Laboratories.



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