Maedi Visna (MV)
seroprevalence
survey 2010

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INTRODUCTION

The aim of this study was to use routinely collected blood samples to evaluate the prevalence of maedi visna (MV) in British sheep flocks. The last large scale seroprevalence survey in GB was carried out in 1995/6 (SAC, unpublished data). The survey involved the testing of 2,277 flocks and 41,593 sheep using bloods that had been collected for routine *Brucella melitensis* surveillance. At that time 1.4% of flocks and 0.19% of sheep were found to be positive for MV in the AGID test. Within the infected flocks the mean number of sheep testing as positive was 13.2%.There was therefore a need for another seroprevalence study to be carried out to estimate the current prevalence of MV in sheep flocks in Britain.

The blood samples used for this survey had been collected by DEFRA for routine annual *Brucella melitensis* surveillance in 2010. From these samples 726 flocks across England, Wales and Scotland were randomly selected to be included in the survey. Only flocks where the owners had given previous permission for the bloods to be used for further testing were included. Twenty samples were collected at random from each flock from sheep greater than 6 months of age (usually adults) or as many as were available if there were fewer than 20 eligible animals in the flock. A total of 11,757 blood samples were screen tested by MV ELISA and the MV AGID test was used to identify individual positive samples.

The report also includes examples of the effects of MV infection in four flocks clinically affected with MV. The number of cases of MV diagnosed in Veterinary Investigation centres in England, Wales and Scotland over the last eight years is also included for interest.

BACKGROUND INFORMATION ON MV

MV is a classed as a lentivirus and the disease has a long incubation period. The clinical signs seen include weight loss, mastitis, pneumonia, arthritis and hind-limb paralysis. Antibodies are produced in response to infection but they do not eliminate the virus. Infected sheep therefore remain lifelong carriers of the virus. There is no cure or vaccine available and infection is often fatal. Infection is mainly spread by aerosol droplets through close nose-to-nose contact but the virus can also be found in colostrum, milk, semen and in-utero infection can occur. The non-specific signs of the disease mean that infection can go undetected in a flock for years and it is often only once the infection level reaches 50% within a flock that clinical signs of infection are seen. As well as causing deaths, flock productivity is affected due to loss of body condition and consequential lower fertility, smaller, less viable lambs being born, poorer quality colostrum, lower milk yields and slower lamb growth rates.

The prevalence of MV is high in countries in southern Europe, such as Italy and Spain. In the north east of Spain a random survey identified MV infection in 97% of flocks sampled. From the 233 flocks tested 40.7% of the sheep were positive for MV on serology (Lujan and others 1993). A serosurvey in Italy, involving 203 flocks, identified 71.4% of flocks as being infected with MV virus (Salvatori and others 2002). There is also a significant level of infection in North America, where the disease is known as ovine progressive pneumonia (OPP). In Canada the seroprevalence of MV infection was estimated to be 19% (Simard and Morley 1991). A survey in the United States found 48% of flocks with evidence of infection, however, the survey was voluntary and open to bias (Cutlip and others 1992). It is thought that the prevalence in northern European countries is at a lower level and in Norway, for example, the disease is notifiable. In 1994, a serosurvey in Finland detected MV in 1.6% of flocks (Sihvonen and others 1999).

SAMPLING & TEST METHOD

A total of 11,757 blood samples from 726 flocks across Scotland, England and Wales that had been collected in 2010 by DEFRA for the purposes of routine *Brucella melitensis* screening were included in the MV seroprevalence survey. Flocks were sampled in each Animal Health region proportionate to the number of flocks in that region in the 2010 Agricultural Survey. In each case, a random selection of 20 sheep from the flock were sampled or all sheep were sampled if there were fewer than 20 in the flock. Adult sheep were generally sampled, although in smaller flocks younger animals greater than six months of age were sampled. MV accredited flocks would also have been included.

The samples were tested in pools of four by an indirect MV ELISA based on a method which has been validated by SAC Veterinary Services to detect 100% of AGID positive samples. This testing procedure is used for the Premium Sheep & Goat Health Scheme MV accreditation scheme routine flock tests. Any pools that tested as positive or inconclusive in the ELISA were tested as individual samples in the MV AGID test.

Prevalence values for samples and for flocks were generated, along with 95% confidence intervals. The results were compared against the values from a previous British prevalence study, using T-tests for continuous variables (comparisons of within-flock prevalences) and chi-squared tests for categorical variables (comparison of number of positive and negative flocks). Chi-squared tests were also used to determine whether there was any statistically significant association (P-value<0.05) between MV presence and month or season of testing, flocksize, housing and presence and number of other animal species on the holding. Both Minitab and Epi-info were used for the analysis, and ArcGIS was used to generate a map to display the differences between regional flock level prevalences.

RESULTS

A total of 87 samples from the 11,757 samples tested were positive for MV in the AGID test. Between 1 and 20 samples were collected from each flock, with 69% of flocks having 20 samples collected (Table 1). This is an individual animal seroprevalence of 0.74% (95% CI 0.59-0.89). Twenty of the 726 flocks tested had one or more AGID positive samples (Table 2). This is a flock seroprevalence of 2.8% (95% CI 1.6-3.9).

No. samples per flock	No. positive flocks	No. negative flocks	Total flocks	% of flocks positive
20	17	485	502	3.39
19	0	4	4	0
18	0	6	6	0
17	0	5	5	0
16	0	3	3	0
15	0	8	8	0
14	0	6	6	0
13	0	7	7	0
12	0	13	13	0
11	0	6	6	0
10	0	15	15	0
9	0	17	17	0
8	0	9	9	0
7	1	16	17	5.88
6	0	16	16	0
5	1	18	19	5.26
4	1	18	19	5.26
3	0	19	19	0
2	0	30	30	0
1	0	5	5	0

Table 1: Number of samples collected per flock

No. of positive samples	No. of flocks	Percentage of total flocks
0	706	97.2%
1	8	1.1%
2	4	0.6%
4	3	0.4%
5	1	0.1%
9	1	0.1%
14	1	0.1%
15	1	0.1%
16	1	0.1%

Table 2: Number of positive samples per flock

The percentage of samples that tested as positive in the infected flocks ranged from 5% to 80% with a mean of 24% (95% CI 13.4-34.5) (Table 3). The number of breeding ewes estimated to be infected with MV in the positive flocks ranged from one to 390 (Table 4).

% prevalence within flock	No. of positive flocks
5%	5
6%	0
10%	4
11%	0
13%	0
14%	1
15%	0
20%	4
25%	2
30%	0
35%	0
36%	0
45%	1
70%	1
75%	1
80%	1
Mean prevalence (%)	24%

Table 3: Prevalence of MV in positive flocks

Positive flock ID	% positive samples	Estimated no. of MV infected breeding ewes in the positive flocks
A	5	18
B	5	20
C	5	10
D	5	7
E	5	37
F	10	74
G	10	13
H	10	3
I	10	17
J	14	1
K	20	1
L	20	160
M	20	27
N	20	n/a
O	25	200
P	25	1
Q	45	40
R	70	390
S	75	98
T	80	320

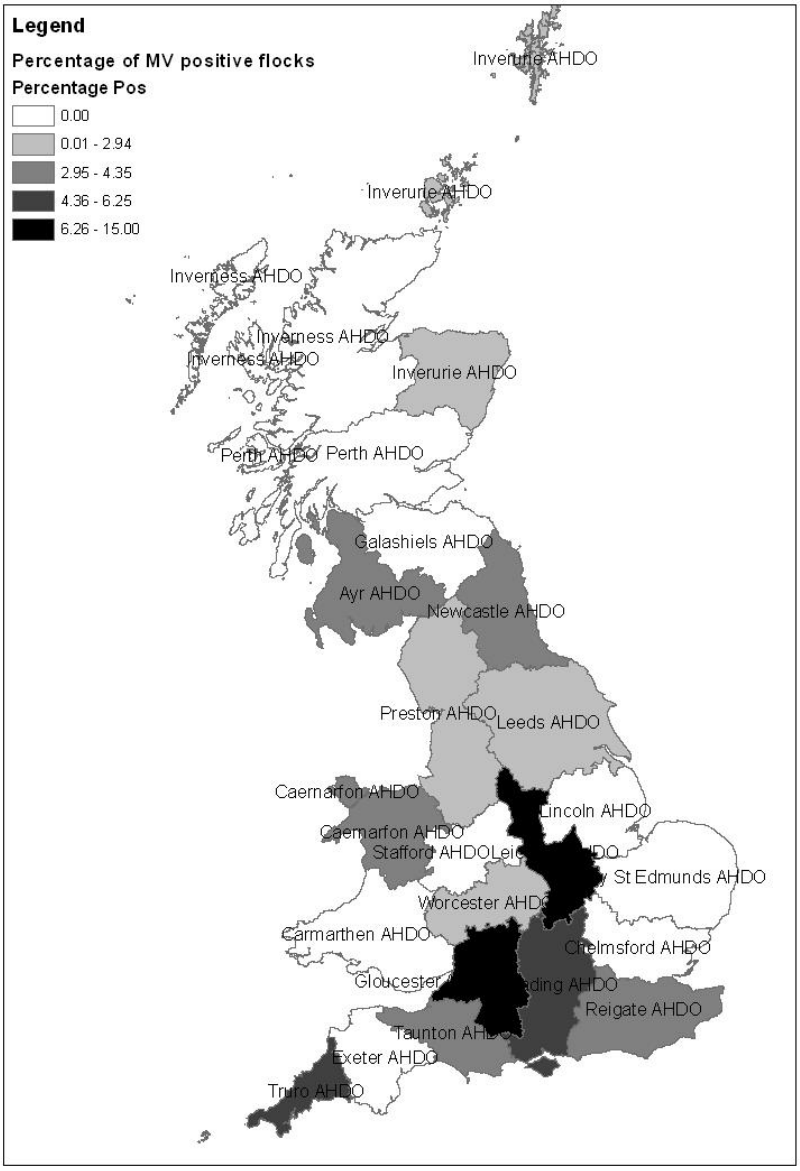
Table 4: Number of ewes estimated to be infected with MV within each positive flock

The number of breeding ewes present in the flock was associated with MV presence in flocks, with a significant linear trend of increasing risk present (Table 5, Chi-squared test for linear trend *P*-value 0.041).

No. of breeding ewes	No. of positive flocks	No. of negative flocks	Percentage positive flocks
Not known	0	4	0.00%
<20	4	226	1.74%
20-50	1	112	0.88%
51-100	1	68	1.45%
101-300	6	136	4.23%
301-500	3	63	4.55%
501-1000	5	68	6.85%
>1000	0	29	0.00%

Table 5: Comparison of MV presence and number of breeding ewes in flock

The prevalence results in each region showed that Leicester~~shire~~ AHDO and Gloucester~~shire~~ AHDO had the highest prevalences (14 and 15% respectively) (Figure 1, Table 6). No distinct spatial pattern of results was observed, with positive flocks present in a



wide range of regions.

Figure 1: Chloropleth map of Animal Health Divisional Office (AHDO) regional flock prevalence.

AHDO Region	No. positive flocks	No. flocks tested in region (% positive)
Ayr	1	25 (4.00%)
Bury St Edmunds	0	10 (0%)
Caernarfon	3	69 (4.35%)
Carmarthen	0	93 (0%)
Chelmsford	0	6 (0%)
Exeter	0	40 (0%)
Galashiels	0	19 (0%)
Gloucester	3	20 (15%)
Inverness	0	58 (0%)
Inverurie	1	39 (2.56%)
Leeds	1	51 (1.96%)
Leicester	4	28 (14.29%)
Lincoln	0	8 (0%)
Newcastle	1	26 (3.85%)
Perth	0	24 (0%)
Preston	1	62 (1.61%)
Reading	1	18 (5.56%)
Reigate	1	26 (3.85%)
Stafford	0	29 (0%)
Taunton	1	25 (4%)
Truro	1	16 (6.25%)
Worcester	1	34 (2.94%)

Table 6: Flock level results for Animal Health Divisional Office (AHDO) regions

COMPARISON OF 1995/6 & 2010 SURVEY RESULTS

There is evidence of an increase in the seroprevalence of MV in the last 15 years. There has been an almost fourfold increase in the individual seroprevalence from 0.19% to 0.74% (Table 7), which was a significant difference (Chi-squared test, $P<0.001$). The percentage of infected flocks has also significantly increased, the value doubled from 1.4% to 2.8% (Chi-squared test, $P=0.015$). The mean percentage of positive samples in infected flocks has almost doubled from 13.2% in 1995/6 in infected flocks to 24% in the positive flocks in 2010 (Table 8, 2-sample T-test, $P=0.049$).

In June 1995, the number of breeding ewes in the national flock was 20.488 million. At a seroprevalence of 0.19% this would estimate the number of MV infected ewes to have been 38,927 at that time (Table 7). By June 2010 the number of breeding ewes in the national flock had fallen to 14.740 million. The seroprevalence of 0.74% detected by this study would indicate that there were 109,076 MV infected ewes in the national flock in 2010.

	1995/6 survey	2010 survey
Individual prevalence	0.19	0.74
Flock prevalence	1.4	2.8
Mean prevalence within infected flocks	13.2	24.0
Estimated no. of infected ewes in national flock	38,927	109,076

Table 7: Comparison of 1995/6 and 2010 survey results

% prevalence within flock	No. of positive flocks – 1995/6 survey	No. of positive flocks – 2010 survey
5%	17	5
6%	1	0
10%	2	4
11%	1	0
13%	1	0
14%	0	1
15%	3	0
20%	2	4
25%	1	2
30%	1	0
35%	1	0
36%	1	0
45%	0	1
70%	0	1
75%	1	1
80%	0	1
Mean prevalence (%)	13.2%	24%

Table 8: Comparison of prevalence of MV infection in positive flocks between 1995/6 and 2010 surveys

DISCUSSION

As the flocks were randomly selected for the surveys, MV accredited flocks were also included. As these flocks are free of infection the level of MV infection in non-MV accredited flocks will be higher than that estimated by the survey.

From the survey results there is evidence that larger flocks have an increased risk of being infected with MV. This could just be due to having more animals so there is more risk of becoming infected or it could be that there is a greater risk of introduction of MV either due to their replacement policy or from neighbouring flocks. Flocks with more than a hundred breeding ewes are at most risk but very large flocks (more than 1000 ewes) appear to be at a lower risk of being infected. The latter may be due to very large flocks being more likely to be closed flocks and that they may be more likely to be hill flocks so have less contact with neighbouring flocks. MV accredited flocks are often smaller flocks and they may be influencing the lower level of infected flocks in those with less than 100 ewes.

Whilst many vets in practice have never diagnosed a case of MV it is likely that infected ewes are not being detected. There are a few possible reasons for this. One is that many of the clinical signs of MV are not specific and so ewes losing condition will often be culled without investigating the reason for the weight loss. Similarly ewes dying from pneumonia may have been suspected to have died due to a Pasteurella infection but MV could be the underlying problem.

There is often little veterinary involvement with sheep flocks. In addition, with MV having been rare in the past it may not initially be considered as a cause of poor condition in a sheep flock. To highlight this point, two heavily infected flocks that were investigated had high levels of bacterial mastitis and it was not initially realised that there was an underlying MV problem. It is likely that one of the main reasons that MV in infected sheep is undetected is that clinical signs are often not seen in a flock until it is heavily infected. By the time over 50% of the flock is infected and MV is diagnosed it is then very difficult to control the infection.

The MV accreditation scheme has been in existence for around 30 years and in that time it has helped keep up to 3,200 members flocks free of infection. SAC Sheep & Goat Health Schemes took over the running of the MV scheme from MAFF in 1996 and until 2010 the average number of accredited flocks to break down with MV infection was 0.1% per annum (Synge and Ritchie 2010). In 2010, there was an increase in the number of breakdowns in accredited flocks to 13 (0.5% of the membership), in 2011 there were seven (0.28%) and in the first five months of 2012 there have been six breakdowns. The majority of routine flock tests take place during the summer months. The risk of having a breakdown with MV has been calculated as being 2.3 times higher if there is also a non-MV accredited flock on the same holding (Synge and Ritchie 2010). This suggests that there is increasing pressure on MV accredited flocks from non-MV accredited sheep.

The availability of MV accredited rams will also have kept many commercial flocks free of infection, for those that are closed flocks other than for the entry of rams. However many commercial flock owners have taken no action to prevent the introduction of MV into their flocks.

In the last few years there have been increasing reports of commercial flocks being found infected with MV and suffering from the clinical effects of infection. Examples of four of these are given.

EXAMPLES OF FOUR FLOCKS WITH CLINICAL MV

Flock A

This was a recently purchased milking flock of 80 Friesland ewes. In the first year under new ownership the scanning figures had been 220% and at the end of lambing the flock performance was 180%. The following year 115 lambs were born to the 122 females that had been bred (94% flock performance figure at lambing). About 25 lambs died in the first few days of life. A ewe which died at the time was diagnosed with Pasteurella pneumonia. The flock had been moved to a new holding and the new parlour was not ready until three months after lambing. By this time there had been a high incidence of chewed teats and a high level of bacterial mastitis. It was also noted that lamb growth rates were poor. When they did start milking through the parlour the milk yield was found to be about a quarter to half of that expected. About eighty lambs were weaned from the 122 females put to the ram (66%). The flock owner also reported that there had been a number of three to four year old ewes drying off unexpectedly and rapidly losing condition. Fifteen ewes were culled after lambing and a further 25 were culled in the autumn due to poor condition and/or mastitis. There had also been a case of visna in the flock with one ewe showing signs of hind limb paralysis. About eight to ten ewes died from pneumonia. Blood sampling of the flock found that 80 out of the 95 adult sheep were positive in the MV AGID test (84%). There was no option but to cull the flock due to the low productivity.

Flock B

This is a flock of 1500 Masham mule ewes. In 2009 there was a problem with single bearing ewes being in poor body condition at lambing. Feeding was adequate and there was no evidence of Johne's disease or of a trace element deficiency on blood testing. Further blood testing revealed that four out of six ewes tested were antibody positive for MV. That year the lambing percentage was lower than expected. About 30 to 40 ewes were separated off for extra feeding due to being in thin body condition and some of these died. There were increased numbers of smaller, weakly lambs born and the ewes had lower milk yields. This resulted in slower lamb growth rates and lower weaning weights. The following breeding season there was a high barren rate though some of this may have been related to the dry summer in 2009 and less grass being available. The flock owner reported that it had been difficult to fatten cull ewes in recent years and that there had been a higher replacement rate.

The flock owner suspected that MV infection had possibly been bought-in in 2001. Due to Foot & Mouth Disease restrictions he had been unable to source replacements from the usual four or five flocks in the north of England and had purchased 300 mule gimmers locally. These had originated from the north of England as ewe lambs. The group had not performed well and there had been high levels of mastitis. There had been problems with poor body condition and rapid loss of condition. There had been a high mortality rate and a high culling rate. Most of them would have been culled within two to three years of arriving. Another possible source of infection could have been a nearby flock as he was aware that all

of the rams in it had tested as positive for MV. In the summer of 2010 the flock owner estimated that there had been a 20-40% reduction in flock productivity and that MV had cost in the region of £30,000-£50,000.

Flock C

This is a flock of 1000 mule ewes and about 300 Blackface ewes. The owner was not aware that they had a high level of MV in their flock. In the previous few years there had been a problem with mastitis and it was only after sampling a proportion of the ewes that it was found that MV was the underlying problem. The results of the blood tests suggested up to 80% of the ewes, and a similar proportion of the stock rams, were infected. Over the previous three years there had been significant problems with mastitis. About 70-100 ewes were culled each year as a result of bad udders. There was also high ewe mortality with about 100 ewes dying a year. These involved ewes with mastitis, others that had lost condition and some with signs of pneumonia. Joint-ill has been a problem in young lambs. MV could be affecting the udders resulting in poorer quality and a lower volume of colostrum, which is pre-disposing the lambs to joint-ill.

Due to such a high level of MV infection in this mule flock, and with the significant losses as a result, the only practical option is to cull the mule flock and to replace it with a cleaner flock over time. It is not economically feasible to cull the whole flock at the one time so it will be phased out over about three years. At the same time an isolated, clean flock will be built up through breeding their own mule replacements from a newly established flock of Blackfaces, which have been tested for MV.

Flock D

MV was diagnosed in a 500 mule ewe flock following a post-mortem examination in early 2012 after a ewe showed signs of inco-ordination. It was one of six to be showing signs of hind limb weakness and inco-ordination at that time. This was the visna form of MV. There had been eight cases of suspected visna between autumn 2011 and the spring of 2012 in relatively young sheep, often one to two crop ewes. In the two months since lambing this year about eight ewes have had one or two swollen knees due to arthritis. This is non-responsive to treatment and progressively worsens, typical of MV arthritis. They have had to be culled for welfare reasons.

There has been an increase in ewe mortality in the flock in recent years. It was 6% in 2010, 8% in 2011 and it was at 5% after just the first four months of 2012. This compares to a median ewe mortality figure of 3-4% over the same period for flocks in the Sheep Animal Health Planning System (SAHPS). There are no other significant health problems in the well managed flock. Replacements are bought in from flocks in the north of England.

Random blood sampling of 48 ewes identified 62% as positive for MV in the AGID test. Almost a third of the positive sheep were four years old. Out of the 17 four year old sheep that had been tested 16 were positive (94%). A similar level of infection was found in homebred sheep as was identified in the bought-in sheep. In retrospect it is thought that the first case of visna had probably occurred in the flock about ten years previously and there had been occasional cases since then until the sudden increase towards the end of 2011.

The costs of the increased mortality in this flock from the start of 2010 to the end of April 2012 have been calculated as £10,210. The breakdown is as follows:

Lost income from adult culls:

Year (size of flock)	No. adult sheep that died	Approx value each	Total lost income	If 3% flock mortality- adult sheep deaths	Approx value to flock if 3% mortality
2010 (400 ewes)	27	£40	£1080	12	£480
2011 (450 ewes)	37	£40	£1480	14	£560
To end 2012 (500 ewes)	23	£40	£920	3	£120
Total lost income			£3480		£1160

Replacement costs due to mortality:

Year	No. replacements	Approx cost	Total cost	If flock mortality 3%	Approx cost if 3% mortality
2010	25 females 2 rams	£105 £500	£2625 £1000	12 females	£1260
2011	36 females 1 ram	£115 £500	£4140 £500	14 females 1 ram	£1610 £500
To end April 2012	23 females	£115	£2645	4 females	£460
Replacement costs due to mortality			£10910		£3860

Carcase disposal costs:

Adult sheep disposal cost = £15

Adult sheep mortality January 2010 to end April 2012: 87 @ £15 = £1305

Adult sheep mortality if 3% per annum as a typical flock for same period: 31 @ £15 = £465

Total cost due to adult sheep mortality in flock: £15,695

Total cost due to adult sheep mortality in typical flock with 3% mortality: £5485

Total cost due to increased mortality from January 2010 to April 2012: £10, 210

This is a very basic costing and does not take into account any costs due to lost production as a result of ewes being in poorer condition or treatment or increased labour costs.

VIDA STATISTICS

Further evidence for an increasing level of MV in the national flock comes from the VIDA (Veterinary Investigation Disease Analysis) statistics (Table 9, Figure 2). These are diagnoses of MV from samples or carcases submitted to disease surveillance centres in England, Wales and Scotland.

Year	No. of clinical cases of MV in VIDA
2004	3
2005	3
2006	0
2007	0
2008	5
2009	3
2010	9
2011	8

Table 9: Clinical cases of disease recorded in VIDA

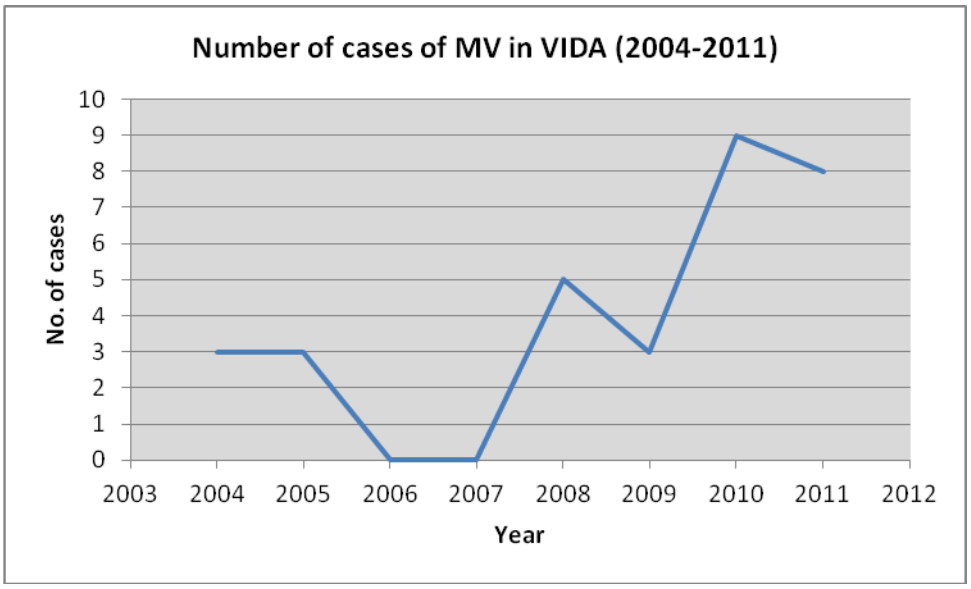


Figure 2: Number of cases of MV diagnosed in Veterinary Investigation Centres

CONCLUSION

The MV survey has shown that there has been an almost four fold increase in the number of sheep that are estimated to be infected with MV compared to 15 years ago. There are twice as many infected flocks now as then and within the infected flocks the average proportion of the flock testing as positive has doubled.

Flock owners need to be taking more action to prevent their flocks from becoming infected with MV. This would include buying replacements from MV accredited flocks, quarantining and testing sheep before introducing them to the flock and, if possible, putting in place measures to reduce the risk of infection entering from neighbouring flocks. Regular monitoring of the flock by blood testing a proportion for MV is also recommended because by the time clinical signs of MV are seen in a flock infection is already at a high level making it much more difficult to put in place an effective control programme.

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