

Quarterly report of investigations of suspected exotic diseases

Vesicular disease ruled out

A veterinarian alerted the Investigation and Diagnostic Centre (IDC) to a suspected vesicular disease after observing erosive lesions in a pet four-month-old female kunekune pig. The pig had been admitted to the veterinary clinic earlier that day, recumbent, showing signs of abdominal pain, low temperature, injected mucous membranes, and erythematous skin lesions on the distal parts of all four legs. The veterinarian euthanased the pig later that day, after it started to have seizures, and noticed the oral lesions. Vesicular disease was ruled out by an AsureQuality initial investigating veterinarian (IIV) who examined the animal and determined the oral erosive lesions to be traumatic in origin.

A veterinarian reported mouth and tongue erosions in four dry cows from a mob of 250 on an Otago dairy farm. All four animals had normal rectal temperatures. An IIV found the mouth lesions were thickened, pyogranulomatous erosions affecting the tongue, buccal area or hard palate. No feet or teat lesions were identified. The animals were in good body condition and were eating well. All other stock were unaffected. The last movement of stock onto the property was six weeks earlier when six in-calf heifers arrived. The IIV ruled out exotic vesicular disease on clinical and epidemiological grounds. The lesions were considered typical of actinobacillosis. Their precipitating cause appeared to be trauma either from feeding kale with woody stalks, or from stones ingested with grass silage fed from a pad with a poorly prepared stony surface.

A single dairy cow on a Taranaki farm was found by a veterinarian to have three ulcers on its tongue and nasal planum. An AsureQuality IIV found the animal had a normal temperature, was not lame and there were no lesions on the feet or udder. No other cattle on the farm were affected, and there was no relevant importation pathway. An exotic vesicular disease was ruled out on clinical and epidemiological findings.

Atypical scrapie ruled out

Histological examination by MAF Biosecurity New Zealand's expert reference pathologist of the brain of a sheep that had died after displaying progressive nervous signs, could not rule out atypical scrapie. Fixed brain material sent for immunohistochemistry to the Veterinary Laboratories Agency, Weybridge, UK, had no evidence of PrP immunolabelling, allowing both scrapie and atypical scrapie to be ruled out.

Equine influenza ruled out

A veterinarian contacted the MAF Biosecurity New Zealand (MAFBNZ) 0800 hotline after a client, who had recently collected

Exotic disease investigations are managed and reported by MAF Biosecurity New Zealand's Investigation and Diagnostic Centre (IDC) Wallaceville. The following is a summary of investigations of suspected exotic disease during the period from July to September 2007.

their USA-bred miniature pony mare from Karaka quarantine facility, reported the horse had developed a mild nasal discharge. As part of enhanced quarantine measures instituted since the equine influenza (EI) outbreak in Australia, a nasal swab from this horse (along with 12 others that came in to Karaka from Kentucky, USA on 25 August) was tested for EI by PCR at IDC Wallaceville, prior to release from quarantine. All results were negative. The entire quarantine cohort of 13 horses had been clinically normal during their two-week quarantine at the Karaka facility, apart from one that had transient travel sickness on arrival. Under the direction of the Incursion Investigator, an equine veterinarian visited the miniature horse and found no signs consistent with EI, only a mild serous nasal discharge with no cough or pyrexia. A second deep nasal swab tested for EI by molecular methods at IDC Wallaceville, again gave negative results.

Brucella canis ruled out

A veterinary pathologist contacted MAFBNZ after histological examination of fixed testicular material from a three-year-old dog revealed a fibrinosuppurative orchitis and epididymitis with cellulitis. The dog had presented with incontinence, lumps in the epididymis as well as two scrotal plaques. *Brucella canis* was ruled out after serum sent to IDC Wallaceville tested negative to the *Brucella canis* card agglutination test.

Leishmania spp ruled out

An 11-year-old spayed female Sheltie tested positive for antibodies to *Leishmania* spp during routine pre-export testing to meet Australian import health standards. The immunofluorescent antibody test (IFAT), carried out by IDC Wallaceville but subcontracted to VetPath Laboratories Western Australia, returned a positive titre of 1:128. The dog was born in Texas USA where she lived for four years before moving to New Zealand in 2000. A local veterinarian working under the direction of the Incursion Investigator found the dog to be healthy with no clinical abnormalities consistent with *Leishmania* infection. Routine haematology and biochemistry analyses showed no abnormalities. A subsequent molecular assay on EDTA blood at Acarus Laboratory, Bristol University, UK, was negative for *Leishmania* spp DNA. A second serum sample tested for *Leishmania* spp antibodies by IFAT at Texas Veterinary Medical Diagnostic Laboratory (TVMDL), USA, gave negative results. *Leishmania* IFAT is known to cross-react

with *Trypanosoma cruzi* (Chagas disease) antibodies. *Trypanosoma cruzi* is endemic in parts of the USA including Texas. IFA testing for *T. cruzi* at TVMDL (the *T. cruzi* IFAT does not cross-react with *Leishmania* antibodies) also gave negative results. Repeat testing of both serum samples with a newly implemented *Leishmania* IFAT at IDC Wallaceville gave negative results. The investigation concluded that the initial result was a false-positive reaction caused by a non-specific reaction or a cross-reaction with other components or agents in the blood.

Coccidiomycosis ruled out in an imported dog

A bulldog imported from Arizona, USA, had a non-healing wound since its importation in November 2006. The wound was considered to be a possible infection with the fungus *Coccidioides immitis*. The dog had a low CFT titre of 1:4 at the time of importation, and at the time of writing had negative titres on both the complement fixation and agar gel immunodiffusion tests. The case has been referred to a small animal medicine specialist to attempt to find an alternative diagnosis.

***Rhipicephalus sanguineus* confirmed**

A veterinarian reported a client who had found a tick on the family dog recently imported from Brisbane, Australia. MAF's reference expert identified the tick as a partially engorged female brown dog tick (*Rhipicephalus sanguineus*), which was estimated to have been attached for five or six days. This timeline fits with the tick having been picked up in pre-import kennels where the dog spent the 10 days before departure. The dog is recorded as having been treated with fipronil (spot-on application) seven days before leaving, in accordance with import health requirements, and was checked by an experienced quarantine officer on arrival at Christchurch. IDC has checked import measures and overseen testing and control measures applied to all in-contact cats and dogs. Serum and EDTA blood from the dog was tested by IFAT for antibodies to *Babesia gibsoni* and a molecular test for *Babesia* spp DNA, with negative results. This case appears to highlight the difficulties associated with arrival inspection as a risk mitigation measure, and the efficacy of fipronil as a spot-on application. MAFBNZ border standards group is examining dog import inspection and treatment procedures, and following up with Australian authorities. *Rhipicephalus sanguineus* has caused temporary infestations in North Island houses on three occasions and is the most commonly intercepted tick. It could establish, especially in northern parts of the North Island, or in heated houses (where temperatures are suitable) in other parts of the country. *Rhipicephalus sanguineus* is not known to have established in New Zealand.

Avian influenza and Newcastle disease ruled out

A MAFBNZ Quarantine Officer alerted the IDC to a foreign pigeon captured from a vessel originating from Zhoushan, China.

Four pigeons had landed on the vessel during bad weather five days before the vessel entered the Nelson port. The approximate position of the vessel when birds landed was 240 nautical miles west of New Caledonia and 600 nautical miles east of Australia. The captured bird was euthanased and a postmortem undertaken at IDC Wallaceville. The bird had a low grade hepatitis with multiple foci of inflammation in which heterophils and occasional histiocytes were prominent. Several sections of trematode worms were present in the small intestinal lumen. Newcastle disease and avian influenza were ruled out subsequent to negative PCR tests on a range of tissues. The other three pigeons were not recovered.

A hobby poultry keeper contacted MAFBNZ after noting increased mortality through autumn in his flock of about 50 birds. Ten to 15 birds died during this period following several days of anorexia, lethargy and dropped wings. A specialist avian veterinarian conducted a postmortem on two birds and considered erythroblastosis (a disease in the leucosis complex) to be the underlying cause. Samples from two other birds tested negative for coccidial oocysts and intestinal nematode eggs. Unfortunately, no material was fixed for histology. Since the initial report in early May, only one further bird had died by the time the investigation was closed in late July. Highly pathogenic notifiable influenza and velogenic Newcastle disease were ruled out on clinical, pathological and epidemiological grounds.

MAFBNZ was contacted after school children found about 15 dead and dying house sparrows in their playground. AnASUREQuality field officer was contracted to collect the affected birds and euthanase any still alive. Necropsies and histology on a full range of tissues from six birds at IDC Wallaceville were unremarkable. Samples tested by real time influenza A and Newcastle disease (ND) RT-PCRs were negative for ND. One duplicate from each pool gave a suspicious result for influenza A. Further testing using H5, H7 and nested PCRs, to eliminate the possibility of low viral RNA yield in the samples, gave negative results for both samples. Samples were also negative after two passages for virus isolation. No *Salmonella* species were isolated from pooled liver and intestines.

Avian pneumovirus ruled out

As part of routine export testing carried out at IDC Wallaceville, five of 30 25-week-old chickens tested positive to avian pneumovirus (APV) using the IDEXX ELISA. None of the birds exhibited signs of respiratory disease that might suggest infection with APV. APV was ruled out following negative immunofluorescent antibody tests (NVSL, Ames, IA, USA).

Infectious bursal disease ruled out

A flock of layers was investigated following detection, through routine infectious bursal disease (IBD) surveillance, of two birds with positive IBD VNTs. Titres were low (1:64 and 1:16). There was no clinical evidence of IBD in the flock. Additional sera were collected from all sheds on the property and 20 young sentinel

birds were introduced into the shed containing VNT positive birds. The introduced birds subsequently tested negative to IBD by PCR. Serological investigation of the flock revealed that 37% of a further 200 birds were positive to the IBD ELISA but only 1.4% were positive to the VNT. The serological results were not consistent with IBD infection in the flock or vaccination. A project is underway at the IDC Wallaceville to develop a more specific VNT for IBD.

A specialist poultry veterinarian reported to MAFBNZ a cluster of cases involving broilers with bursal atrophy, increased mortality and cellulitis. Swabs and tissue specimens were submitted to the IDC to rule out an exotic viral aetiology. Cloacal swabs and spleen were negative on PCR testing for IBD. Newcastle disease was ruled out on PCR tests of the liver, spleen and kidney. A cytopathic virus detected by virus isolation in the bursa and spleen of one bird was identified by electron microscopy as an adenovirus. It tested negative by haemagglutination inhibition (HI) test for type III adenovirus – the cause of egg drop syndrome. This adenovirus is therefore likely to be type I or type II. Histopathology of the lung, liver, spleen, bursa and thymus was not conclusive for inclusion body hepatitis. Examination of the mortality rates for all sheds in the production system indicated that the mortality was increased in the first week and from 28 days onwards. This trend was consistent in the last two production runs but not in the production figures prior to April 2007. Mortality figures include culls, dead birds and moribund birds. Two farmers interviewed perceived the largest category of increased mortality to result from leg problems, mainly femoral head necrosis, a condition of chickens and turkeys that may be associated with several different bacterial infections such as staphylococci, *E coli* or streptococci. Predisposing factors for femoral head necrosis include immunosuppressive viruses such as IBD virus, chicken anaemia virus and non-infectious bone pathologies such as hypophosphataemic rickets. The combination of an adenovirus isolation from the spleen and bursa, postmortem reports of small bursas and splenomegaly, and increased prevalence of femoral head necrosis and cellulitis, does indicate that an immunosuppressive condition may be involved. Further work will be undertaken to type the adenovirus and improve the sampling strategy to define the condition.

Avian blood parasites investigated

A pathologist reported suspicious intracytoplasmic inclusion bodies in a blood smear from a New Zealand crimson rosella. The blood smear was submitted as a routine check before the bird was admitted to Auckland Zoo. Review of the smears by Massey University pathologists ruled out blood parasites. A staining artefact may have been present as no inclusion bodies were observed in a second blood smear made several days after the first.

Tracheal mites ruled out

A beekeeper reported the loss of one to three hives per apiary in about 18 apiaries. The interviewing apiary officer suspected

starvation and robbing of the hives as the most likely cause of hive loss. However, as a precaution, bees from three hives were tested for tracheal and external mites (including *Tropilaelaps* species). No tracheal mites were found in any of the 22 bees dissected. Varroa mites and a small number of other mites were found: *Tyrophagus communis* (mould mite – Acaridae, one mite), *Carpoglyphus lactis* (dried fruit mite – Carpoqlyphidae: Acari, two mites), and *Melittiphis alvearius* (melittiphis mite – Laelapidae: Acari, one mite). These other mites are of no pest significance to honey bees; they are scavengers on waste products in the hive (pollen, honey, fungal growths). All are regularly found in samples from MAFBNZ's apiculture surveillance programme.

Gobio sp investigated

A vial apparently containing two small fish preserved in formalin was delivered to IDC Tamaki with no accompanying paperwork, return address or identification of sender. IDC Wallaceville sent the fish to NIWA to the Marine Invasives Taxonomic Service (MITS), which identified *Gobiopterus semivestita* (syn *G semivestitus*). This species is a small, tropical freshwater fish from northern Australia and Papua New Guinea and was first found by the Department of Conservation in a small tributary of the Ngunguru River (near Whangarei, Northland) several years ago. Its origin in New Zealand is unknown; it may occur here naturally and been undetected, it may have arrived in New Zealand via natural dispersal or it may have been introduced via shipping. As this species is reportedly already established in New Zealand and has been here for several years, no further action was required

Exotic oyster ruled out

A member of the public reported an unusual looking oyster growing on the hull of their recreational vessel moored at Gulf Harbour Marina, Whangaparaoa, Auckland. The vessel had not been moved in the past six months. The report was emailed to the Ministry of Fisheries and passed on to the Animals and Marine Incursion Investigation Team. A specimen sent to the MITS was identified as the Pacific oyster *Crassostrea gigas*. Its unusual shell morphology was probably the result of growing in a low energy (few waves or little current) environment. As this species was introduced to New Zealand many years ago and is well established and widely farmed commercially, it is of no biosecurity concern.

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