



PROJECT REPORT

AI viruses in migratory shorebirds and nomadic waterfowl in South Australia Final Project Report 01 September 2009 – 14 May 2010

The project, “Avian Influenza viruses in migratory shorebirds and nomadic waterfowl in South Australia”, was granted funding by DAFF in October 2009. Project progress is presented, following the guidelines for the report as set out in Section 8 of the Funding Deed of the same name, between the Commonwealth of Australia, PIRSA (Grantee) and Flinders Partners Pty Ltd (FP). The report contains:

- (a) a description of Project progress during the period covered by the Report, against the Objectives, Milestones and Key Performance Indicators;
- (b) an explanation of why any activity, outcome or Milestone specified in this Agreement for the period covered by the Report was not conducted or achieved;
- (c) a description of how Activities, Objectives or Milestones not conducted or achieved are to be conducted or achieved;
- (d) financial statements for the receipt, holding, expenditure and commitment of the Funding during the period covered by the Report;
- (e) a report of all Project Material and all Intellectual Property in Project Material created or arising during the period covered by the Report; and
- (f) a report of all Assets created or acquired during the period covered by the Report.

Project Objectives

- To seasonally monitor for the presence of avian influenzas in migratory shorebirds that travel to South Australia in large numbers from AI infected zones in Asia.
- To enable South Australia to develop and refine wild bird surveillance techniques for AI, as well as to promote interaction between animal health and wildlife personnel.

Proposed Activities

- Sampling from the migratory shorebirds as they arrive (September) at the austral summer feeding habitat (Gulf St Vincent and Coorong) in southern Australia.

- Undertake preliminary analysis to determine AI status of all birds sampled.
- Undertake follow-up detailed analysis of any positive samples to identify the type of AI involved.

(a) a description of Project progress during the period covered by the Report, against the Objectives, Milestones and Key Performance Indicators

Outcome	Comment	Key Performance Indicators	Comment	Milestones	Comment
Targeted sampling of wild bird populations for avian influenza viruses.	Attempted sampling occurred on 17 and 18 December 2010, with the assistance of the South Australia Wader Study Group. The team visited beaches in the south eastern areas of the state, around the Port MacDonnell area. Unfortunately the team was unable to catch any birds, in spite of there being migratory wader species present on the beaches visited. Another attempt at catching waders was made from 1 to 5 March 2010. Both the South Australian and the Victorian wader study groups were involved in the trapping. Trapping was done as part of routine banding work done on migratory wader species in southern Australia. Successful catches were made on both 2 and 4 March 2010, from beaches around the Port MacDonnell area the south east of the state.	FP Collection of up to 300 cloacal samples from relevant migratory shorebird species during the period September 2009 to March 2010, in accordance with the national strategy for wild bird surveillance	Successful catching and cloacal sampling occurred on 2 and 4 March 2010. A total of 351 cloacal swabs were taken from various species of migratory wader birds, including Red Necked Stints, Sanderlings, Ruddy Turnstones, and a Curlew sandpiper.	1. Provide DAFF Project Officer and Australian Wildlife Health Network (AWHN) with a detailed plan of surveillance activities and results, which are to be entered into eWHIS database by early May 2010.	Results so far have been communicated to the AWHN and DAFF. Final results will be communicated as soon as they are available
Identification of virus sub types from samples collected.		Forwarding of samples to the PIRVIC laboratory at Attwood (Victoria) for laboratory testing, with identification down to virus subtypes from positive samples	All samples were sent to PirVIC laboratory at Attwood (Victoria) for avian influenza testing. Swabs were pooled into pools of 3. Initially, 6 pools were positive. Further testing demonstrated that only 3 swabs were positive for Influenza Type A. The species involved were 2 Red Necked Stints and one Sanderling.	2. Completion of progress report and acceptance by the DAFF Project Officer by 15 October 2009. All available avian influenza data entered in eWHIS and demonstrated 30 October 2009.	Progress report completed
Communication of test results		Communication of test results in a format accessible to scientific colleagues	As soon all testing is completed, the results will be communicated to the Australian Wildlife Health Network (AWHN).	3. Provision of monthly reporting of sampling and results to the Australian Wildlife Health Network and quarterly reporting to the Wild Bird Avian Influenza Surveillance Steering Group from the activities in Schedule 1 of this Agreement.	Reports provided to the AWHN, both monthly and quarterly.
				4. Provision of a final report to the DAFF Project	Report provided (attached)

Officer by 9 May 2010 and report acceptance by DAFF by 14 May 2010. All available avian influenza data entered in eWHIS and demonstrated 21 June 2010. Appendix 1 for results)

(b) an explanation of why any activity, outcome or Milestone specified in this Agreement for the period covered by the Report was not conducted or achieved;

None necessary

(c) a description of how activities, Objectives or Milestones not conducted or achieved are to be conducted or achieved;

None necessary

(d) financial statements for the receipt, holding, expenditure and commitment of the Funding during the period covered by the Report;

Attached (Appendix 2)

(e) a report of all Project Material and all Intellectual Property in Project Material created or arising during the period covered by the Report; and Project Material

Not applicable.

(f) a report of all Assets created or acquired during the period covered by the Report.

APPENDIX 1

Results of Migratory Wader Sampling in south eastern South Australia 2010:

Date Sampled	Species	No. samples	Type of samples	Place sampled (Near Port MacDonnell, SA)	Results (AIV PCR-initial)		
					<i>Positive</i>	<i>Negative</i>	<i>Pending</i>
2/03/2010	Red Necked Stints	91	cloacal	Danger Point	5 pooled samples positive (no specific species)	100 pools of 3	0
	Sanderling	204	cloacal	Danger Point			
	Ruddy Turnstone	1	cloacal	Danger Point			
	Curlew Sandpiper	1	cloacal	Danger Point			
	Unrecorded	3	cloacal	Danger Point			
4/03/2010	Red Necked Stints	29	cloacal	Nene Valley	1 pooled sample positive (no specific species)	17 pools of 3	0
	Ruddy Turnstone	22		Nene Valley			

Initial Positive Samples – repeat test results:

Pool/Swab No.	Sample No.	Species	Virology					Repeat Full AIV PCR	Comment
			AHV Culture	AIV H5 PCR	AIV H7 PCR	AIV H Type	AIV SEQ		
20	58	Red Necked Stint	-	-	-	-	-	Negative	
20	59	Red Necked Stint	-	-	-	-	Influenza A	POSITIVE	no bands for sequencing. Therefore we are re-extracting and testing on HA Subtyping PCR again
20	60	Sanderling	-	-	-	-	-	Negative	
29	85	Sanderling	-	-	-	-	Influenza A	POSITIVE	no bands for sequencing. Therefore we are re-extracting and testing on HA Subtyping PCR again
29	86	Red Necked Stint	-	-	-	-	-	Negative	
29	87	Sanderling	-	-	-	-	-	Negative	
37	109	Sanderling	-	-	-	-	-	Negative	
37	110	Sanderling	-	-	-	-	-	Negative	
37	111	Sanderling	-	-	-	-	-	Negative	
62	184	Sanderling	-	-	-	-	-	Negative	
62	185	Sanderling	-	-	-	-	-	Negative	
62	186	Sanderling	-	-	-	-	-	Negative	
68	202	Red Necked Stint	-	-	-	-	-	Negative	
68	203	Sanderling	-	-	-	-	-	Negative	
68	204	Sanderling	-	-	-	-	-	Negative	
101	301	Red Necked Stint	-	-	-	-	-	Negative	
101	302	Red Necked Stint	-	-	-	-	-	Negative	
101	303	Red Necked Stint	-	-	-	-	Influenza A	POSITIVE	no bands for sequencing. Therefore we are re-extracting and testing on HA Subtyping PCR again

Methodology

The Victorian and South Australian Wader Study Groups were responsible for catching the migratory waders in this project. They provided their assistance as part of their routine banding activities. They used a canon-netting technique, where birds are observed on an area of beach, and then a canon net is set up and camouflaged. The group then retreats off the beach, keeping the net in sight. The net is deployed when there are sufficient wader species in the catching zone. Birds are gently extracted from the net, taking care to protect them from the elements while they are in the net (keeping them dry and shaded) and placed into shaded, protected holding pens.

Birds are then individually measured, weighed and banded (and some have electronic trackers attached). They are then sampled by gentle cloacal swabbing. Birds are then released.

Cloacal swabs are placed immediately into a viral transport medium (as supplied by the laboratory), labelled with date, location and species, and are kept cool (refrigerated at 4°C). Once all samples are obtained, they are packaged up into a cooled container and are transported overnight to the laboratory (PirVIC), where they undergo testing.

Conclusion:

We can consider the migratory wader population around the south eastern coast of South Australia as one population, consisting of a few different species, as evidenced by the fact that they spend time together on intertidal zones.

The migratory wader birds that arrive in South Australia visit various areas in Asia. Recent Wader Study Group results from geolocators attached to four Ruddy Turnstones have shown that these birds fly non-stop from beaches in Victoria and South Australia to Taiwan, where they spend a couple of weeks feeding. They then fly northwards through eastern Asia to Siberia, where they breed. They return to southern Australia in spring, where they are often found on the same beach as the previous year (*Clive Minton, pers comm.*)

A total of 351 birds were sampled, of which 3 were confirmed positive for Influenza A virus on PCR. No further sequencing was possible on these samples. All other samples were negative on PCR testing (348 samples).

This is an estimated apparent Avian Influenza Virus prevalence of 0.0085 (0.0029 – 0.0248), using test sensitivity of 90% and specificity of 99%.

As this was the first time that migratory wader species have been sampled for AI viruses in South Australia, it enabled PIRSA and University of Flinders' staff to gain valuable experience in wader handling and sampling techniques, as well as establishing valuable relationships and interaction between Biosecurity Animal Health staff, University of Flinders personnel and members of the Wader Study Group. Project objectives one and two were thus both achieved.

APPENDIX 2

Total Expenditure for project

Laboratory Costs:	Lab name:
	PirVIC – Attwood
Cost for testing- AI PCR:	\$35.00
No of samples	351
No. of visits - August to March:	2
Cost per positive sample:	\$207.90
Total no. of positive samples (initial and final)	18 (6 pools of 3)
Total lab costs for testing cloacal samples:	\$12,285.00
Total lab costs for positive samples:	\$3,742.20
Courier costs (for samples to lab and supplies from lab)	\$2,020.70
TOTAL Laboratory cost:	\$18,047.90
Other costs:	
Salary per person (PIRSA)	\$2,500.00
Number of PIRSA people required:	3
PIRSA Vehicle costs per day	\$60.00
No. days PIRSA vehicle required:	7
PIRSA Vehicle cost for 7 days:	\$420.00
Km travelled	2376
Fuel and Maintenance costs	\$1,734.48
Total traveling costs	\$2,154.48
Wader Study Group Payment	\$750.00
Total Staff costs	\$7,500.00
TOTAL Other Costs:	\$10,404.48
Commonwealth Contribution:	\$5,000.00
SA State Contribution:	23,452.38
TOTAL Proposed expenditure:	28,452.38