



Field observations and assessment of the response to an outbreak of White Spot Disease (WSD) in Black Tiger Prawns (*Penaeus monodon*) farmed on the Logan River in November 2016.

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21 February 2017

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## **Executive Summary**

This report provides independent documentation and analysis of events related to a White Spot Disease (WSD) outbreak in Black Tiger Prawns (Penaeus monodon) cultured on the Logan River from late November 2016 until February 2017. Disease was first observed on the index farm (1IP) on 22<sup>nd</sup> Nov 2016 and spread rapidly to affect multiple ponds adjacent to and downwind from the index pond, suggesting on-farm spread via aerobiological means (aerosols) and probably via mechanical vectors (insects, toads, birds). By Monday 5<sup>th</sup> Dec WSD had spread to a second farm (3IP), 1 km north of the index farm and the White Spot Syndrome Virus (WSSV) was found in wild prawns (*Metapenaeus* spp., Acetes spp., n = 6) sampled from the Logan River near 3IP. By Thursday 8<sup>th</sup> Dec WSD had spread to a third farm (4IP) around 2 km downriver, while a separate compartment of the first farm (2IP) recorded clinical disease on 12<sup>th</sup> Dec. An isolated case of a single clinically normal mud crab (Scylla serrata) positive for WSSV was recorded in the outlet canal of a 7<sup>th</sup> (then non-infected) farm (7ARP, 7.3 km east of 1IP) on 23<sup>rd</sup> Dec. Subsequently, disease spread to a 5<sup>th</sup> farm around 5 km downriver (5IP) with clinical disease recorded on 28th Dec. Spread between these 5 farms did not appear random. In all cases index ponds at each farm were located at the southern ends of intake canals, downwind from the mainly northerly winds at the time. Location of index ponds along intake canals was the only consistent risk factor once other risk factors (PL source, pond stocking date, food source, water quality) were assessed at each site. Index pond location and non-random distribution of crustaceans and vectors within the intake canals suggests that the affected farms bought in the disease agent through their intake canals via unidentified, possibly planktonic, carrier hosts. The sixth farm infected (8IP) was drawing water from southern Moreton Bay and was positive for WSSV in samples taken on 24<sup>th</sup> Jan 2017 during the later stages of harvest. The final farm (7ARP) remained uninfected until 11th Feb 2017. This farm may have been infected from nearby 8IP where hundreds of birds were observed wading in WSSV positive ponds on 3<sup>rd</sup> Feb. The WSSV positive P. monodon sampled from the river near 3IP (25th Jan) and on mud flats near the outlet of 8IP on 8th Feb 2017 are considered likely to be farm escapees, but genetic testing is required to confirm this assumption.

While its possible that introduction of WSSV to farms 4IP, 5IP and 8IP downriver from the 1IP/2IP/3IP farm cluster may be explained by predictable downstream movement of water and/or carriers as per CSIRO modelling, the mode of introduction of the virus into index farm 1IP and the anomalous positive mud crab in the outlet of 7ARP requires thorough investigation. Sources of feed, equipment or hatchery supplies of PL do not appear to explain the emergence of the disease at 1IP. Instead, the epidemiology and chronology of disease spread together with evidence of significant recreational fishing effort in and adjacent to the intake canal at 1IP, strongly suggests, in my professional opinion, that the incursion pathway was most likely introduction of WSSV via the 1IP intake canal. Indeed, surveys by Fisheries officers allegedly found several groups of recreational fishers using imported green prawns as bait within 500 meters of the intake of farm 3IP, and of these 33% of bait samples were positive for WSSV. This pathway is plausible given evidence that; 1. Increasing numbers of recreational fishers are using imported prawns as bait, and 2. Biosecurity breakdowns at the international border resulting in c. 50-54% of imported green prawns sold at the retail counter being WSSV positive in the leadup to Christmas/New Year 2016. The risk profile for this pathway may have increased since the 2009 Import Risk Analysis for prawn products, meaning that the risk analysis needs to be thoroughly reviewed and updated to more accurately reflect the various risk pathways and new emerging diseases prevalent in the world today.



Keywords: White Spot Disease, WSSV, Black Tiger Prawn, Mud Crab, Logan River, Moreton Bay

## Abbreviations, acronyms and definitions

#### **Abbreviations and Acronyms**

AAHL Australian Animal Health Laboratory (Geelong, Victoria)

**ALOP** Appropriate level of protection APFA Australian Prawn Farmers Association

ARP At risk property/premise Biosecurity Queensland BO circa/approximately c.

CT value Cycle threshold value (for qPCR).

**DAWR** Department of Agriculture and Water Resources

dps days post stocking IΡ Infected property/premise

National Association of Testing Laboratories, Australia **NATA** 

OIE Office International des Epizooties, the world organisation for animal health

PL Post larvae QLD Oueensland

Polymerase Chain Reaction (a genetic diagnostic test) PCR qPCR Quantitative PCR (also known as real time PCR)

RA Risk analysis South Australia SA WA Western Australia WSD White spot disease

White spot syndrome virus WSSV

1IP First infected property (Index prawn farm)

Second infected property (2<sup>nd</sup> prawn farm nearest index prawn farm) 2IP Third infected property (3<sup>rd</sup> prawn farm, c. 1 km north of the index farm) 3IP

Fourth infected property (4<sup>th</sup> prawn farm, c. 2 km downriver from the index farm) 4IP 5IP

Fifth infected property (5<sup>th</sup> prawn farm, c. 7.3 km downriver from the index farm) Seventh infected property (7<sup>th</sup> prawn farm, c. 9 km east of the index farm) Eighth infected property (8<sup>th</sup> prawn farm, c. 10 km east of the index farm) 7IP 8IP

9IP Ninth infected premise (a retail bait and tackle shop, c. 2 km south west of the index farm) 6ARP Sixth at risk property (a 6<sup>th</sup> prawn farm, c. 11 km east of the index farm, that was non-operational)

#### **Definitions**

CT value

Cycle threshold value for quantitative PCR. A positive reaction using quantitative PCR (also known as real time PCR) is detected by accumulation of a fluorescent signal. The CT (cycle threshold) is defined as the number of PCR reaction cycles required for the fluorescent signal to cross a threshold that exceeds background levels of fluorescence. The CT value is inversely related to the level of target genetic material in the sample. A high level of virus (in the case of WSSV) means fewer cycles are required to cross the threshold target, resulting in a lower CT value (typically <20 for a strong positive). A low level of target genetic material in the sample requires more cycles to generate a positive result (potentially up to 40 cycles or more) to exceed the threshold and be regarded as a positive result. Samples where the threshold value of fluorescence is not reached during the duration of the test are classified as negative results.

Index pond/farm

The individual animal identified as being the first to contract a contagious disease during a disease outbreak is called the index case. The pond in which the first diseased animal is located is therefore called the index pond, while the first farm where the first index case was detected is called the index farm.



## 1.0 Introduction

White spot disease (WSD) is an internationally notifiable disease of crustaceans caused by White Spot Syndrome Virus (WSSV), a double-stranded DNA virus of the genus Whispovirus within the family Nimaviridea (OIE 2016). Previous WSSV incursions into Australia include its detection in broodstock prawns (*Penaeus monodon*) and mud crabs (*Scylla serrata*) fed frozen imported prawns at an aquaculture hatchery in Darwin Harbour in December 2000. In that case wild mud crabs and prawns adjacent to the hatchery outlet were also transiently infected with WSSV, but over time this infection fizzled out and subsequent testing indicated that the virus did not become established in Darwin Harbour (East et al. 2004, 2005).

The current WSSV incursion was first reported in Black Tiger Prawns (*Penaeus monodon*) cultured on a prawn farm (1IP) taking water from the Logan River, SE QLD on 22<sup>rd</sup> November 2016. Biosecurity Queensland (BQ) was alerted, obtained diagnostic samples, and a confirmed diagnosis of White Spot Disease (WSD) was obtained from the Australian Animal Health Laboratory (AAHL) on 1 December 2016. The Office International des Epizooties (OIE) was provided with an immediate notification on the same day, and on the morning of 2 Dec 2016, DigsFish Services was engaged with the objective of visiting the affected farm to document proceedings and provide updates to the fisheries and aquaculture industries regarding what was happening on the ground. At the same time information on the response was recorded with the objective of updating industry resources and the relevant Aquavetplan manuals at a future date, samples were taken for storage onsite for future industry based research and biosecurity advice and assistance was provided to farm staff, the prawn farming industry, and Government authorities upon request.

Upon confirmation of WSD at the first farm, BQ immediately enacted an eradication program based on Aquavetplan (Department of Agriculture 2013) involving destruction and decontamination of affected farms, a program that is ongoing at the time of publication. The virus proved to be highly contagious for *P. monodon*. By Monday 5th Dec disease had spread to a second farm (3IP), 1 km north of the index farm and the virus was also detected in a small number of wild prawns from the Logan River (*Metapenaeus* spp., n = 5, *Acetes* spp. n = 1). Disease subsequently spread in a non-random fashion to a fourth farm (4IP, clinical disease 8<sup>th</sup> December), a separate compartment of the first farm (2IP, clinical disease 12th Dec) and a fifth farm c. 5 km downstream (5IP, clinical disease 28th Dec 2016), all of which were drawing water from the Logan River (Figure 1). Infection of Mud Crabs (Scylla serrata) was also confirmed in drains of several infected farms, as well as an anomalous detection of one mud crab sampled on 23 rd Dec 2016 in the outlet canal of a seventh uninfected farm (7ARP) around 7.3 km east of the index farm (Figure 2). A sixth farm (8IP) was positive for WSSV in samples taken on 24th Jan 2017 during the later stages of harvest, and clinically diseased prawns were noticed on the last remaining farm (formerly 7ARP, now 7IP) on 11 Feb 2017. On-farm activities undertaken by DigsFish Services director Dr Ben Diggles (BKD) from 2 Dec 2016 were documented at the time in field notes that formed the basis of a series of nine Industry Situation Reports (Appendices 1-9) that were published on a near immediate basis in order to facilitate rapid flow of information from the on-farm situation to the wider prawn farming and fishing industries. The present report summarises the field observations and analyses outcomes from those Industry Situation Reports, and provides some suggestions for consideration when updating the relevant Aquavetplans and the Import Risk Analysis for prawn products.

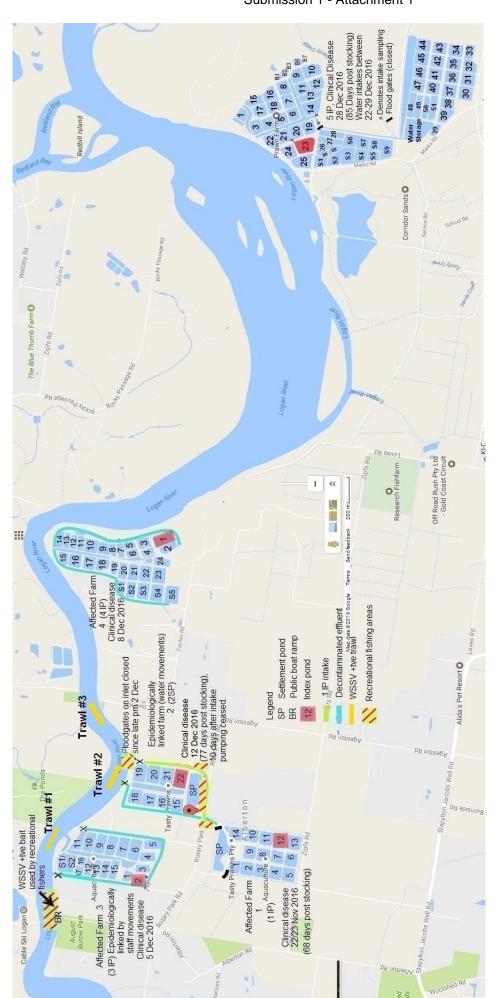


Figure 1. Map showing location of the first 5 affected prawn farms (1IP to 5IP), with first dates where clinical disease was observed in index ponds (red shading) and other relevant data.

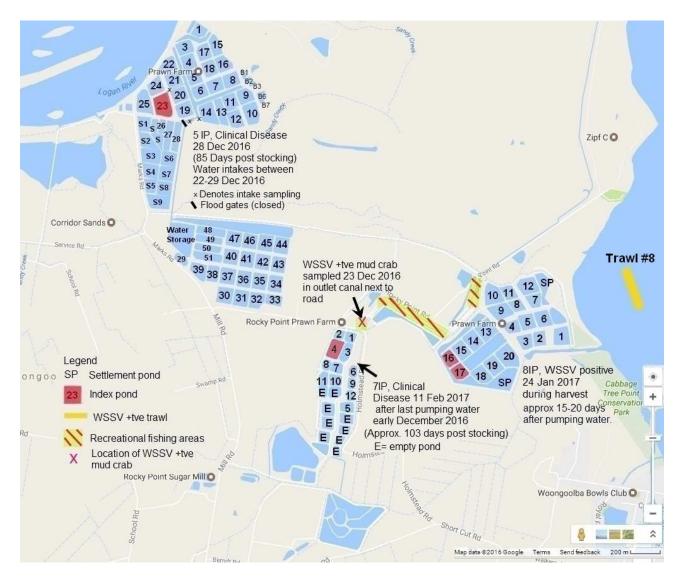


Figure 2. Map of east Logan River prawn farms showing location where a WSSV infected mud crab was sampled in the outlet canal of the then uninfected 7ARP, in an area regularly frequented by recreational fishers. Pond 4 at 7ARP (now 7IP) exhibited WSD on 11 Feb 2017, 8 days after several hundred birds were observed wading in the drained WSSV positive ponds 16 and 17 at 8IP.

## 2.0 Materials and Methods

The information contained in the series of nine Industry Situation Reports was obtained through direct interviews with farmers on WSD affected farms, as well as from observations, samples and measurements taken by BKD on-farm and pond side during the biosecurity response. Information published by BQ and the Chief Veterinary Officer of Australia during notifications to the OIE are also included where necessary to provide a comprehensive chronology of the sequence of events and a better understanding of the broader ramifications of the disease outbreak. This information will be useful for farmers who need to consider their options going forward, including consideration of alternative production strategies for farm operation, which may hinge largely on whether WSSV becomes established in the environment, or not.

## 3.0 Results

### 3.1 DigsFish Field Notes

The dates of various notable events recorded during interviews with Logan River prawn farmers, commercial prawn fishers working on the Logan River, and BQ staff as well as personal observations by BKD during farm visits from 2 December 2016 until Tuesday 14<sup>th</sup> February 2017 are presented in Table 1. Details of field samples taken (Table 2) and water quality data for the pond containing the index case at 1IP (Table 3) are also included. For more details of field observations between 2 December and 13<sup>th</sup> January 2017, including eyewitness accounts, readers are referred to the complete series of 9 Industry Situation Reports presented in Appendices 1-9 at the end of this report. Details of key events that occurred between 13<sup>th</sup> January and 14<sup>th</sup> February 2017, which are presented at the end of Table 1, are new and previously unpublished.

Table 1. Events leading up to and during a White Spot Disease (WSD) outbreak in Black Tiger Prawns (*Penaeus monodon*) cultured on the Logan River from late November 2016 to February 2017.

Date	Location	Observations/comments/references
Mid/late May 2016	Logan River	Strong wild Banana Prawn run comes to unusually abrupt halt during onset of colder weather in mid/late May 2016, unprecedented in previous 20 years (Lee Dreyer, commercial prawn fisher, personal communication to BKD, 11 Dec 2016)
13-27 Sept	Prawn farm 3IP	Ponds stocked with post larvae from Rocky Point hatchery, starting with ponds 2 (index pond) and 3 (13 Sept), then pond 1 (14 Sept), pond 6 (15 Sept), ponds 4 and 5 (16 Sept), ponds 14 and 15 (19 Sept), pond 7 (20 Sept), ponds 11, 12 and 13 (21 Sept), pond 8 (22 Sept), pond 16 (23 Sept), pond 10 (26 Sept), and ponds 9 and 17 (27 Sept).
14-27 Sept	Prawn Farm 1IP	Ponds stocked with post larvae from Rocky Point hatchery. Ponds 5 and 6 stocked first (14 Sept), ponds 12 (index pond) and 13 stocked on 15 Sept, then ponds 10 and 11 (19 Sept), 3 and 4 (20 Sept), 1, 2, 7, 8 (21 Sept), Pond 9 (22 Sept), and pond 14 (27 Sept).
26-28 Sept	Prawn farm 2IP	Ponds stocked with post larvae from Rocky Point hatchery. Ponds 21, 22 (index pond) and 18 stocked first (26 Sept), followed by 17 (27 Sept) and 16, 19 and 20 (28 Sept).
Mid Sept	Prawn farm 4IP	All ponds stocked in 1 week with post larvae from own hatchery starting from northern ponds (8 to 20) first then southern ponds (7 to 1 (index pond)).
23-30 Sept	Prawn Farm 5IP	Ponds in northern eastern section of farm stocked with post larvae from own hatchery. Ponds 1, 15, 16 stocked 23 Sept, ponds 3, 17, 18 on 24 Sept, 4, 5, 6 on 26 Sept, 7,8,10 on 27 Sept, 9, 11, 13 on 28 Sept, 12, 14, 19 on 29 Sept, 20 and 21 on 30 Sept.
4-5 Oct	Prawn Farm 5IP	Ponds in northern western section of farm stocked with post larvae from own hatchery. Ponds 22, 23 (index), 24, 25, 27 stocked on 4 Oct, ponds 26 and 28 stocked 5 Oct.
31 Oct	Prawn farm 2IP	Pond 15 stocked with post larvae from Rocky Point hatchery.
22Nov	Prawn Farm 1IP Index case	BKD subsequently advised by farmers that prawns (4 gram <i>P. monodon</i> ) in pond 12 (stocked 15 Sept 2016, 68 days post stocking) were off feed, lethargic, swimming in circles near surface at pond edges. Feed rates

Date	Location	Observations/comments/references
22 Nov		significantly reduced. Feed for previous 2 weeks is Ridleys extruded
		pellets that are cooked at 85-95°C for 30-45 minutes (Ridley 2016, see
		Appendix 10) after starting on CP extruded pellets (Thailand). Pond water
		quality data leading up to the disease event is presented in Table 3.
23 Nov	Prawn Farm 1IP	BKD subsequently advised by farmers that clinical signs continue and
		minor mortalities (2 or 3 dead prawns) noted today in pond 12.
24 Nov	Prawn Farm 1IP	BKD subsequently advised by farmers that prawn mortalities had increased
		in Pond 12 based on observations at 6 am. Biosecurity QLD notified of
		problem by phone at 9 am. Farmers collect and take samples to BQ
		laboratory for diagnostic testing. White spots on carapace noted at time of
		sampling in some diseased prawns and laboratory technician advised of
		this when handing samples over for testing. Farmers observe bird activity
25.34	D E 11D	increasing (c. 120 seagulls observed).
25 Nov	Prawn Farm 1IP	BKD subsequently advised by farmers that BQ staff arrived and took 20
		live prawns from pond 12 to the laboratory for further testing. Farmers
		advised the visiting BQ staff that the prawns in pond 12 had white spots.
		Advice to farmers from BQ e-mailed that afternoon was that the disease
		was not <i>Penaeus monodon</i> Mortality Syndrome (PMMS). Farmers advised verbally to do whatever was required to get best result from that pond.
		Pond flushing commenced.
26/27 Nov	Logan Area	Prevailing north easterly winds 15 knots. Light/moderate rainfall 26/27 <sup>th</sup>
20/27 1107	Logun 7 neu	Nov (57 mm at Logan River Water Treatment Plant 26 <sup>th</sup> Nov) results in
		increased likelihood of discharge from cane farm drains into 1IP outlet
		canal and then into Logan River (Figure 4).
28 Nov	Prawn Farm 1IP	BKD subsequently advised by farmers that mortalities continued to
		increase in pond 12. Farmers very concerned and take fixed water samples
		to Bribie Island Aquaculture Research Station for testing for toxic algal
		species. Results are negative for toxic algae.
29 Nov	Prawn Farm 1IP	BKD subsequently advised by farmers that mortalities continued to
		increase in pond 12 and now clinical signs of disease were observed in
		adjacent ponds 11 and 13. In the morning of 29 <sup>th</sup> Nov farmers advised
		verbally over phone by BQ they could drain harvest. Samples of diseased
		prawns from ponds 11 and 13 taken by farmers to BQ laboratory for
		diagnostic testing. While at laboratory farmers were advised verbally by
		BQ staff that samples from pond 12 were being prepared for histology, and
		that they had white spots but they looked more like spots due to calcium
		deficiency. By that afternoon advice from BQ changed and farmers were
20.14	D E 170	verbally informed to isolate ponds 11, 12 and 13. Pond flushing ceased.
30 Nov	Prawn Farm 1IP	BKD subsequently advised by farmers that the BQ Laboratory verbally
		advised them of preliminary positive result for WSSV from pond 12
		samples. Results were 5/10 prawns positive using one qPCR platform,
		0/10 positive using a second platform. Verbal advice from BQ staff was to
		chlorinate the affected pond ASAP. Farmers were unable to acquire the
		required amount of chlorine without a permit, but purchased and
1 Doc	Prawn Farm 1IP	administered what they could that day.
1 Dec	riawii Faliil HP	BKD subsequently advised by farmers that clinical signs of disease now spreading to 7 pends (12, 13, 11, 7, 8, 10 and 5). WSSV positive (±tve)
		spreading to 7 ponds (12, 13, 11, 7, 8, 10 and 5). WSSV positive (+tve) results confirmed by AAHL (CT values range 14.0–21.13). Biosecurity
		QLD emergency powers declared and OIE alerted to detection and advised
		intent is to respond, contain and eradicate the disease from the affected
		farm and confirm freedom in nearby farms and in wild crustaceans. The
		source of the outbreak is unknown. Earthen bank bulldozed to form bund
		Source of the outbreak is unknown. Earthen bank bundozed to form bund

Date	Location	Observations/comments/references
1 Dec		to block outlet from the 1IP settlement pond, but 1IP outlet canal remains
		open to Logan River. Northerly winds and light rainfall occurs in area.
2 Dec	DigsFish Services	BKD alerted to WSD outbreak 7.40 am and DigsFish Services engaged to
		visit the affected farm to document proceedings, update fisheries and
		aquaculture industries, take notes with objective of updating industry
		resources and the relevant Aquavetplan manuals, take samples for future
	Prawn Farm 2IP	industry based research, and provide biosecurity advice if required.  BKD conducts inspection of second farm (2ARP) and finds exit of outlet
	(then 2ARP)	canal from 1IP flows past 2ARP and is interconnected to 2ARP via the
	(then 27 tree)	latter's settlement pond. BKD observes that exit to 1IP outlet canal still
		open to river allowing ingress/exit of any crustaceans in the outlet canal
		into/out of river. Farmer at 2ARP advised by BKD to cease pumping water
		from river after he discloses pumping water that morning. He advises that
		no one told him that pumping water from the river may be risky. No bird
		control officers on site. Wind 10-15 knots North East in afternoon.
	Prawn Farm 1IP	BKD observes clinically diseased prawns in Ponds 13, 12, 11, 10, 8, 7, 5
		and 4. Many hundreds to thousands of mortalities evident at pond edges in
		ponds 11 and 12 with dead and moribund prawns displaying reddish tinge,
		loose carapace, and around 25% of dead prawns displaying clinical signs of white spots on cephalothorax or abdomen (Figure 3). Prawns in ponds 1,
		2, 3 appear healthy, status of 9 and 6 uncertain. Bird control officers on site
		and birds now under control, but large numbers of flying insects observed
		at pond edges. Chlorination of pond 12 commenced at 4.45 pm using
		liquid chlorine (nominal 10-13% v/v sodium hypochlorite) transported to
		the site in a 13,000 litre tanker and applied to various positions around the
		pond periphery (target>30 mg/L free chlorine for >24 hrs, peak of 50 mg/L
		achieved). Mortality of remaining prawns occurs within 2-3 hours of
	T + 1 1 1 1TD	application of the chlorine.
	Intake canal to 1IP	Exiting 2ARP onto Rotary Park Rd at 2.30 pm BKD observes recreational
		fishers fishing in the 1IP intake canal from the roadside. BKD advised by fishers they are castnetting for prawns to take to other locations as live bait.
		Fishers also advise they and many others regularly frequent the canal for
		line fishing and cast netting. Biosecurity QLD officers at the 1IP control
		point verbally advised by BKD of the fishing activity in the control zone.
3 Dec	Phone hookup	Phone hookup with BQ, APFA, DigsFish for shared understanding of how
	Î	to respond. Local Control Centre established at Coopers Plains. BKD
		advised by BQ that 3 ponds now chlorinated at 1IP (11, 12, 13) and that
		1IP outlet canal downstream from bund will be blocked off at entrance to
		river and treated with hydrogen peroxide (0.2%) ASAP. Restricted area to
		be declared in the river for commercial and recreational fishers. Heavy
		storm hits Logan region that evening increasing likelihood of discharge from cane farm drains into 1IP outlet canal and then into Logan River
		(Figure 4).
4 Dec	Prawn Farm 1IP	1IP outlet canal blocked off from river and treated with hydrogen peroxide
	outlet canal	(approx 0.2%) on or around 4 <sup>th</sup> Dec.
5 Dec	Tackle shop (later	At 9.30 am BKD inspects bait prawns at local tackle shop – they appear
	designated 9IP)	apparently normal. BKD advised by shop staff that some bait prawns on
		sale are received from local trawlers working the Logan River.
	Prawn Farm 3IP	10 am, arrived at 3ARP, BKD advised by farmers that diseased prawns
	(then 3ARP)	observed that morning in pond 2. They last pumped water pm of 4 Dec as
		no one had told them that pumping water from the river may be risky.
		Indeed, they felt they were operating in an information vacuum and had no

Date	Location	Observations/comments/references
5 Dec	Location	advice or help at all from government authorities. BKD observes 5-6 dead
3 Dec		prawns and clinically diseased prawns swimming in circles at pond edges.
		Feed is Ridley extruded pellets. No bird control officers on site. BKD goes
		to control point at 1IP (750 m to south) and reports suspect WSD in second
		farm. Returns to 3 ARP at 11 am to find 3ARP staff already have blocked
		off both outlet canals with earthen bunds. Staff advise 2 dead crabs
		observed in intake canal recently. Samples of prawns taken from 3ARP by
		BQ on 1 Dec were taken from feeding trays, and are all expected to be
		negative for WSSV. However cast netted samples from pond 2 that
		afternoon had clinical signs of WSD. Biosecurity QLD advised by BKD
		that feed tray sampling likely to have much reduced sensitivity for
		detection of diseased prawns and advise cast net sampling. BKD helps
		farm staff to set up footbath and handwash facilities at farm entrance. Wind
		12-15 knots North /NW in afternoon.
	Intake canal to 1IP	6.10 pm, BKD observes no advisory signage evident or controls over
	munic cuntil to 111	movement of public or fishing activities near road adjacent to 1IP intake
		canal.
	Prawn Farm 1IP	BKD observes that ponds 11, 12, 13 treated with chlorine. Settlement pond
		partially treated with chlorine (100,000 L). Clinically diseased prawns
		observed swimming in circles at pond edges in pond 10 with heavy
		mortalities evident (100's of prawns visible along pond edges). Occasional
		mortalities becoming evident at pond edges in ponds 9, 8, 7, 6, no major
		changes in ponds 5 and 4. Ponds 3, 2, 1 and 14 appear normal. Large
		numbers of canetoads observed moving around ponds at night.
	Logan River	5 apparently normal Metapenaeus sp. (from 60 collected, prevalence =
		8.3%) and 1 Jelly Prawn ( <i>Acetes</i> spp.) subclinically infected with WSSV
		are collected by beam trawl adjacent to 2IP (Trawl #2 n = 4 <i>Metapenaeus</i>
		sp.) and 1IP intake (Trawl #3 n = 1 <i>Metapenaeus</i> sp., 1 <i>Acetes</i> spp.). All
		CT values range between 37.5 – 39 (i.e. possibly carrier state).
6 Dec	Intake canal to 1IP	Overnight rain, 15 knot northerly wind. At 12.45 pm, BKD observes no
		controls evident over movement of public or fishing activities near road
		adjacent to 1IP intake canal.
	Prawn farm 2IP	BKD inspects all ponds from 12.45 pm, no evidence of diseased prawns,
	(then 2ARP)	relatively low numbers of insects present. Prawns in one pond here (pond
	D E 31D	15) are smaller as they were stocked later (31 Oct).
	Prawn Farm 3IP	BKD informed by farm staff at 9 am that diseased prawns now evident in
	(formerly 3ARP)	pond 3 adjacent to index pond 2, possibly due to intake water or
		aerobiological (aerosol) movement from pond 2 due to strong north-NW
		winds over previous days. At 3.45 pm BKD on farm and abnormal
		behaviour observed in prawns from pond 3 including lighter coloured
		moribund prawns swimming at the water surface, with occasional erratic flicking along the pond edges. At pond 2 WSD had progressed rapidly
		since yesterday morning with significantly higher numbers of dead and
		moribund prawns (3-5 per meter) visible along the pond edges, mainly
		along the upwind (north) pond edges with fewer on the downwind edge.
		Chlorine truck arrives 4 pm with enough chlorine to dose 1 pond only, so
		we choose to dose pond 3 with chlorine to try to stop spread to ponds 4 and
		5. BKD takes BQ staff for a pond inspection and notes they are unable to
		properly observe pond activity (e.g. failing to see large mudcrabs) due to
		lack of aquatic experience and proper equipment (e.g. high quality
		polarized lenses). After 2 hours BKD returns to pond 3 to observe prawns
		dying with large numbers already dead in the downwind (south east) corner
	L	ajing with targe numbers arready dead in the downwind (south east) collect

Date	Location	Observations/comments/references
		of the pond.
7 Dec	Prawn Farm 3IP	BKD advised by farmers over phone that pond 2 was chlorinated at 4 pm, but outlet canals surrounding the farm still not treated, and continuing to fill. AAHL confirms samples taken 5 Dec from pond 2 are positive for WSSV (CT values not reported). Prawns in pond 1 immediately adjacent to index pond 2 still appeared normal. Heavy rain that afternoon (Figure 4).
8 Dec	Prawn Farm 3IP	BKD arrives on farm at 2 pm. No chlorine in drain at this time. At 2.30 pm farm staff advised by BQ that WSSV infected prawns detected in river "near Alberton boat ramp" (see 5 Dec for dates and locations). BKD conducts site inspection and finds no abnormal signs or abnormal prawn behaviour in any ponds (except chlorinated ponds 2 and 3). A bird control officer interviewed by BKD at pond 11 confirmed that no one was conducting bird control at 3IP overnight between 10 pm and 6 am. Chlorination of 3IP outlet canal begins starting in north west corner from around 7.20 pm.
	Intake canal to 1IP	BKD interviews a local resident living in the house opposite the IIP intake about the frequency of fishing activity in the intake. Resident informs BKD that it was not unusual to have 3 to 4 groups of people a week fishing in the intake, many with cast nets, others targeting bream and other estuarine fish species, sometimes right up near the intake pipes. Also informs that around 2 weeks prior, Dept. of Fisheries may have caught some people for illegal fishing in the intake. Another local resident then pulled up to inquire what I was doing and asked whether I had been fishing, showing that local surveillance of fishing activity on the road near the intake had increased, however no signs or warnings to anglers not to fish there had been erected. BKD inspects entrance to 1IP intake and informed by farmer at 2ARP that floodgates had been closed since late pm on Friday 2nd December. Gates configured to allow water to enter the intake canal, but not drain back out, meaning that the intake to IP1 represents a semi-enclosed sub-habitat of the Logan River. BKD assists farmer to close off the overflow pipe with reinforced plastic at around 6.15 pm to prevent water from escaping if it rains. Since the intake has been closed since Friday 2nd December BKD suggests in sitrep report #3 (Appendix 3) it is well worth intensively sampling the inlet to try to improve epidemiological understanding of the origin of the index outbreak.
	Prawn Farm 1IP	BKD informed by BQ staff at 7.30 pm that they had nearly finished dosing all ponds at 1IP so were now positioned to focus more attention on 3IP.
	Prawn Farm 4IP (then 4ARP)	A 3 <sup>rd</sup> farm approximately 2 km downriver from 1IP/2IP reports clinical signs of WSD in ponds 1, and 3 (and possibly 2), on the same day as prawns in ponds 1, 2, and 4 are detected as WSSV positive by BQ (CT values range 20.2-43.04). BKD informed by farmer that they were pumping water from the Logan River up until they saw first clinical signs as no one had advised them of the potential risk of doing so. Water temperature on 5 Dec in index pond 1 was 28.6°C, DO 3.95 mg/L, pH 7.59, salinity 30.8 ppt. "Signs of disease" in pond 3 subsequently thought by farmer to be due to lightning strike during storm of 7 Dec when pond results are negative for WSSV (results supplied on 12 Dec). Water quality data for 4IP does not disclose any significant differences between index ponds and other ponds on site. Each pond behaves in a slightly different manner dictated more by when algae blooms occur etc., rather than being markedly influenced by pond position in the intake (data available upon request). Feed is CP 4003 extruded pelleted shrimp feed (Thailand).

Date	Location	Observations/comments/references
8 Dec	Logan River Area movement controls	Movement controls declared as of 8 December by Department of Agriculture and Fisheries in an attempt to prevent all persons including commercial and recreational fishers from moving all known carriers of WSSV in the area of the Logan River downstream from Jabiru and Luscombe weirs to the river mouth (for map of the closure area see Fig. 2
9 Dec	Prawn Farm 3IP	in sitrep report #4, Appendix 4).  BKD advised by farmer that 5.30 am that morning Pond 4 at 3IP had been chlorinated by unsupervised truck drivers and several other chlorine trucks were parked outside 3IP ready to continue decontamination of non PCR positive, non-clinically diseased ponds at that site. It was suggested during the industry phone hookup that morning that these trucks could be better utilised by directing them to finish chlorinating the 3IP outlet canal or the known infected (PCR positive and clinically diseased) ponds at 4IP.
10 Dec	Epidemiological observations Sitrep4	BKD notes that as for all 3 other infected farms, the index ponds on 4IP showing first clinical signs of WSD (and/or were identified by testing as being WSSV positive first) were those ponds that were furthest away from the intake pumps at the end of intake canals. BKD notes (point 2 in Sitrep report #4, Appendix 4) that "This consistent picture throughout all farms infected so far suggests the spread of the disease between farms is not random or caused by movements of birds or human activities, but instead is highly likely to be associated with pumping in virus (or more likely, hosts infected with the virus) from the river. A plausible hypothesis might be that ponds nearest the water intakes are receiving water that is in the intake/supply canals for a shorter time period, resulting in prawns being exposed to lower viral titres. In contrast, the ponds furtherest from the intakes may be taking water that has higher viral titres due to less water flow concentrating virus and/or vectors (e.g. jelly prawns Acetes spp.) at the very ends of the supply canals where water flow is least and water quality is likely to be reduced".
	Prawn Farm 3IP	Attempt emergency harvest of several ponds. BKD advised by farmers that the emergency harvested ponds were then refilled with recycled water ready for chlorination. However BQ instead chlorinates unharvested ponds filled with harvestable prawns instead.
	Prawn Farm 4IP	Ponds 1, 2, 3, 4, 5 and 6 now chlorinated, together with the outlet discharge canal to the east of the property, resulting in the deaths of large numbers of fish (mainly Sea Mullet, Eels, Yellowfin Bream, and Forktailed Catfish).
11 Dec	Tackle shop 9IP	At 11 am BKD inspected local tackle shop to find them selling fresh locally caught (from Logan River) chilled and frozen prawns despite the movement control area restrictions. Several species of prawns were observed with white spots grossly similar to signs of WSD, including Banana Prawns ( <i>Penaeus</i> sp.), Greasyback Prawns ( <i>Metapenaeus</i> sp.), river shrimp ( <i>Macrobrachium</i> sp.) and palaemonid shrimp. Other potential WSSV hosts on site included live Yabbies ( <i>Callianassa</i> sp.). BKD immediately notifies BQ of potential breaches of movement control restrictions and possible presence of WSSV on premises and collects samples of prawns with white spots. By 2.45 pm two BQ officers are on site interviewing staff and taking samples, including confiscation of samples procured earlier by BKD (Table 2). Some samples collected with gross signs of WSD subsequently test positive for WSSV in the BQ laboratory, but confirmatory testing at AAHL is inconclusive. At 4.30 pm local commercial prawn fisher informs BKD of anomalous abrupt end of wild Banana Prawn run in May 2016, but testing of archived prawn

Date	Location	Observations/comments/references
11 Dec		samples taken from the Logan River by the commercial fisher then stored
		for months in the tackle shop freezer all subsequently test negative for
		WSSV.
	Prawn Farm 3IP	Wind 10 knots east-south easterly. At 12.30 pm BKD observes prawns in
		ponds 1 and 5 that are randomly sampled by castnet ( $n = 60$ per pond)
		appear normal.
12 Dec	Prawn Farm 4IP	At 5 pm BKD observes very high numbers of dead jelly prawns mixed in
		with dead <i>P. monodon</i> in chlorine treated index ponds 1 and 3. Ponds 1, 2,
		3, 4, 5, 6, 7, 8 and 24 now chlorinated as of today. No evidence of clinical
		disease in any other ponds. Large numbers of live jelly prawns observed in
		ponds 9, 10, 12, 15, 17, and S5. BKD notes a pair of grapsid crabs still
		feeding on dead prawns in chlorinated pond 5, over 24 hours after
		treatment. Both crabs were on the dirt bank at the pond edge feeding on a
		dead prawn that flicked out of the water during chlorine treatment, but both
		reentered the chlorinated water upon approach.
	Other tackle shops	BKD informed by shop staff that BQ staff had visited to confiscate locally
	in area	caught prawns held for sale in bait freezers in several tackle shops in the
		Logan River/Jumpinpin area.
	Prawn Farm 3IP	Wind 10 knots East-SE. At 11.30 am BKD begins to sample and fix (95%
		ethanol) prawns in ponds 6 and 7 that are PCR positive for WSSV and
		scheduled for chlorination today. Sample of 60 P. monodon castnetted
		from pond 6 found no gross signs of disease evident. Sample of 60 prawns
		cast netted from pond 7 found 4 prawns (6.6%) showing clinical signs of
		white spots on carapace. Moderate numbers of jelly prawns observed in
		both ponds. Pond 7 is chlorinated starting at 1.45 pm, pond 6 is
		chlorinated starting 3.10 pm. Pond 10 also sampled at 3.20 pm and all
		prawns (n = 60) apparently healthy. By 4 pm in pond 7 both $P$ . $monodon$
		and Jelly Prawns were swimming in an agitated state at the water surface
		due to chlorine exposure. BKD estimates that around 7-10% of the $P$ .
		monodon that were visible were showing clinical signs of WSD with white
		spots on the carapace. At 4.15 pm some emergency harvested cooked
		prawns observed being transported to freezers at 1IP in sealed containers.
	Prawn Farm 2IP	Clinical disease begins to be observed in pond 22 (south easterly end of
	(then 2ARP)	intake canal). These prawns later confirmed to be WSSV positive at 77
		days post stocking (CT values from 17.1-35.1). Feed is Ridleys extruded
		pellets for previous 4-5 weeks after starting on CP extruded pellets
		(Thailand).
	Prawn Farm 4IP	From 4.30 pm BKD takes random samples of 60 <i>P. monodon</i> from ponds 8
		and 18 for fixation and archiving on site in 95% ethanol. Several prawns
		from pond 8 had what the farmer referred to as "redback" syndrome with
		focal bilaterally symmetrical reddish areas along the dorsal cephalothorax
		and abdomen of affected prawns. A low number of prawns also had
		"saddleback" like deformities of the abdominal segment just before the
		telson. Ponds 1, 2, 3, 4, 5, 6, 7, 8, 13, 14, 20, 21, 22 and 24 all now
		chlorinated as of today. After sampling, BKD interviews farmer who had
		just received hard copy documents for the first time that outlined the results
		of testing (from SAN AAHL report 16:03985 and Biosecurity QLD – pond
		1 = 66% positive for WSSV, pond 2 = 3% positive for WSSV, Pond 4 =
		4% positive for WSSV, all other ponds negative) and other information
		explaining what WSD is and the significance of it to Australia (extracts out
		of the Aquatic Animal Diseases Significant to Australia Field Guide (4th
		Ed), BQ information leaflets, etc.). Upon his request, I went through the

Date	Location	Observations/comments/references
12 Dec		documents with him explaining various aspects. The farmer noted that
		provision of this information in a written form much earlier in the response
		prior to destruction activities would have allayed much confusion and
		stress at the farm - people at the coalface in a fluid situation are often the
		last to hear of developments in the response process and therefore regular
		communication and updates (in written form as this provides substance)
		can assist in making response processes more transparent (thus improving
		compliance and reducing stress for those most affected at the coalface).
		The farmer also noted that it would be desirable that turnover of BQ staff
		on the farm is minimised to improve rapport between farmers and
		Government as well as increasing farmer confidence/knowledge of exactly
		who is on their farm at any given time.
13 Dec	Bait prawns in	Sunfish environmental officer informs BKD that WSD-like signs
	general	(calcification deposits) have been observed on bait prawns in the recent
		past in Australia. BKD advises the officer to inform BQ and provide
		samples of any suspected WSD infected prawns in bait supplies.
	Tackle shop 9IP	BKD observes at 8.45 am the owners beginning the process of voluntarily
		decontaminating the yabby aquarium system. BKD also shown by staff
		thawed bait prawns (Metapenaeus sp.) sourced from outside the closure
		area with large numbers of small white spots on the carapace. While the
		signs appeared more likely to represent a storage/processing artefact, under
		the circumstances the decision by tackle shop staff to withdraw them from
		sale is applauded as the correct one. BKD informs BQ to get officers to
		attend the site ASAP to collect the prawns as samples and help
		decontaminate the area. These prawns were subsequently tested by BQ and
		AAHL and were apparently WSSV negative.
	Prawn Farm 2IP	Wind 10 knots easterly. BKD inspects 2IP at 9.45 am, met by farmer and
		the BQ site controller. We proceeded on foot to pond 22 where BKD
		observes clinically diseased <i>P. monodon</i> with signs of WSD swimming on
		the surface of the pond and also moribund in the shallows. There were also
		dead prawns every 1 to 2 meters along the pond edge. There was only 1
		bird control officer on site at the time and as it was clear that the pond had
		WSD and he could not leave it unattended to patrol the remainder of the
		site, BKD asks him to call in extra bird control personnel. BKD samples
		60 prawns for fixation into ethanol using a new castnet under supervision of the site controller. Of prawns sampled 7 of 60 (11.7%) had gross signs
		of the site controller. Of prawns sampled 7 of 60 (11.7%) had gross signs of WSD. BKD noted that index pond 22 was again the pond furthest away
		from the intake pumps and was taking water from the southern end of the
		supply canal (last pumped 2 Dec). BKD notes this consistent picture
		throughout all 4 farms infected so far indicates the spread of the disease
		between farms is not random, or likely to be caused by movements of birds
		or human activities, but instead is highly likely to be associated with
		pumping in virus (or more likely, hosts infected with the virus) from the
		river. BKD observed BQ staff leave 2IP unattended from 11.30 am-1pm.
	Intake canal to 1IP	BKD observes entrance to 2IP and intake to 1IP from 11.30 am till 1.00 pm
		and confirms no BQ field staff on site at 2IP with WSSV infected prawns
		in pond 22 and only 1 bird control officer on site. BKD also observes still
		no controls evident over movement of public or fishing activities in 1IP
		intake for this same time period, nor any advisory signage erected.
		Fortunately, no one is observed fishing in the intake.
	Prawn Farm 4IP	BKD inspects 4IP beginning at 1.10 pm and randomly samples 60
		prawns into ethanol fixative from ponds 10, 11 and 17 between 1.15 pm

Date	Location	Observations/comments/references
13 Dec		and 6 pm. Ponds S1, S2, 15 and 16 had also been treated since 12 Dec,
10 200		meaning ponds 1, 2, 3, 4, 5, 6, 7, 8, 13, 14, 15, 16, 20, 21, 22, S1 and S2
		now chlorinated as of today. BKD observes no evidence of clinical
		disease in any of the 3 ponds sampled on 4 IP today except for some tail
		deformities in ponds 10 and 11.
	Conversation with	BKD is contacted by WA Fisheries and Agriculture staff by phone re: host
	WA Government	range details underpinning their plans for domestic import restrictions on
		uncooked prawns and other potential WSSV hosts/vectors from the Logan
	representatives	
14 Das	Prawn Farm 3IP	River area. BKD provides host range information to caller.
14 Dec		Chlorination of all ponds scheduled for completion by COB today.
	Prawn Farm 2IP	Index pond (22) chlorinated late PM today.
	Supermarkets in the	BKD surveys several Coles, Woolworths and IGA supermarkets in the
	Gold Coast area	Gold Coast area (n = 4) and without exception all are selling green,
		uncooked imported prawns unpackaged, loose by the kg over the retail
		counter without any signage to customers alerting them that they should
		not used them as bait.
		BKD NOTE: No such advisory signage has been observed at any seafood
		counter at any supermarket I have been to in Brisbane, Sunshine Coast or
		Bribie Island between 14 Dec 2016 and 14 Feb 2017.
	Western Australia	WA Department of Fisheries implements import restrictions on
		Queensland prawns and worms to prevent translocation of WSSV into
		WA. In a press release (MR36-16) dated 14 Dec, a departmental
		spokesperson stated "We are moving quickly to reduce the risk of the virus
		spreading here, by restricting the import of all live or uncooked prawns, or
		parts of prawns and polychaete worms, from Queensland," he said.
		"Unless tested and certified free of WSD, none of these products from the
		wild, farmed or processed in Queensland can be imported into Western
		Australia for human consumption or bait"
15 Dec	Federal Govt and	BKD participates in national phone conference providing official update
	Industry phone	for the WSD outbreak situation. One very important piece of information
	conference	that was provided by employees of the Federal Department of Agriculture
	(minutes in	and Water Resources (DAWR) was that between May and December 2016,
	Appendix 11)	of the 448 consignments of imported frozen raw prawns bought into
		Australia, 73 consignments (16.3%) tested positive for WSSV (Appendix
		11). They also noted that 100% of shipments were being tested, but failed
		to mention that several importers supplied their own samples to
		government for testing. BKD noted at the time (sitrep report #6, point 2,
		Appendix 6) that "it will be important to ascertain the exact sampling
		methodologies used by Federal biosecurity and border security staff to
		arrive at these figures, so that the potential likelihood of false negative test
		results in any non-positive consignments can be better assessed."
	Tackle shop 9IP	BKD observes at 4 pm that BQ Field officers have shut down, cleaned out
		and chlorinated all freezers in the tackle shop that held frozen, potentially
		WSSV contaminated locally caught bait prawns, as well as all of the
		recirculation systems that housed live yabbies. Staff were in the process of
		restocking the freezers with bait prawns sourced from outside the fishing
		closure area.
	Alberton Boat	BKD observes at 4.20 pm that there were no advisory signs or fisheries
	Ramp	personnel informing boaters or fishers of the fishing closure conditions at
	,	the Alberton Boat ramp.
	Prawn Farm 3IP	BKD observes at 4.30 pm that all ponds and drains had been chlorinated
		including settlement ponds
	1	

Date	Location	Observations/comments/references
15 Dec	Prawn Farm 4IP	Farmer reports to BKD by phone that chlorination of final 5 production
		ponds was completed by COB today
	Intake canal to 1IP	BKD observes intake to 1IP at 4.30 pm to find still no controls evident
		over movement of public or fishing activities in 1IP intake, nor any
	Prawn farm 2IP	advisory signage erected.  BKD approaches entrance to 2IP and is unable to locate a BQ site
	Trawn farm 2n	controller. BKD, enters the decontamination area, then spoke to the single bird control officer on site, who was placed near the index Pond 22, which was chlorinated yesterday. There were large numbers of dead <i>P. monodon</i> and Jelly Prawns visible along the edges and pond margins. BKD inspects the remaining ponds 21, 20, 19, 18, 16 and 15, and samples 60 prawns from Pond 17 for genetics and left the samples fixed in 95% ethanol (Table 2) inside the feed hut at 7 pm. All prawns in remaining ponds appear healthy.
	Prawn Farm 3IP	Farmer rings BKD at 9 am to inform him that he had just witnessed a live
		prawn swim over to him at pond 1, 3 days after it had been chlorinated. He thought it may have survived by hiding near the inlet boards where a dilution effect may have occurred.
	Prawn Farm 3IP	Chlorination of all drains and inlets of 3IP to be completed today
	Prawn Farm 4IP	Chlorination of all drains and inlets of 4IP to be completed today
17 Dec	Prawn Farm 5IP	Allowed water intake to attempt to correct water quality issues.
19 Dec	Industry Phone hookup	BKD informed some farms downstream from 4IP starting to harvest ponds. APFA advised that some bait samples from the tackle shop (9IP) test positive for WSSV in BQ laboratory (NOTE: later confirmatory testing at AAHL is inconclusive).
21 Dec	Technical Working Group phone conference	Noted by group during phone conference that chlorination of production ponds had been completed on all 4 infected properties. It was also disclosed, that "3 or 4" of the bait prawns sampled from the tackle shop (9IP) had real time PCR CT values of 32-35 in the BQ Laboratory, but histology showed no confirmed inclusions. Subsequent confirmatory qPCR testing by AAHL for these samples were reported as negative, yielding an overall indeterminate result. BKD notes (sitrep report #6, point 7, Appendix 6) that "histopathology of bait samples that had been frozen for possibly 2 or more weeks prior to fixation in 95% ethanol would result in many artefactual changes that may greatly reduce the sensitivity of histopathology as a diagnostic tool under such circumstances, especially with prawns due to the fact that crustacean tissues autolyse very quickly after death".
	Prawn farm 2IP	BKD inspects 2IP at 2.30 pm and confirms with BQ site manager that chlorination of all ponds has been completed. BKD witnesses that chlorination of the settlement pond there was about to commence.
	Prawn farm 1IP	BKD attends a meeting at 1IP with BQ and the affected farmers from 1IP, 2IP, 3IP, and 4IP, where strategies for finalizing decontamination and beginning the process of draining and disposal were discussed. The meeting finished with resolutions from BQ to finalise disposal strategies in close collaboration with farmers. BKD meets a federal investigator from DAWR who was at the premise earlier interviewing prawn farmers.
22 Dec	Prawn farm 5IP (then 5ARP)	Prawns in pond 23 test negative for WSSV. Farm has water quality problems and to avoid mass prawn mortalities begins taking in a total of approximately 54,450 tonnes of water at the top of the tide over next 7 days from this date until 29 December. Without water treatment this is a high risk activity as CSIRO particle tracking modelling showed that WSSV

Date	Location	Observations/comments/references
22 Dec		particles from upstream sites in the Logan River are likely to have been
		spread past this location by this date (Figure 5).
23 Dec	South Australia	Leon Bignell, MP, Minister for Agriculture, Food and Fisheries declares a White Spot Disease Notice enacted under Sect 37 of SA Livestock Act 1997, declaring a livestock standstill in relation to decapod crustaceans (Order Decapoda) and polychaete worms (Class Polychaeta) for the purpose of controlling or eradicating White Spot Disease. The notice prohibits the entry or importation into South Australia of "live or dead crustaceans of the Order Decapoda, including but not limited to prawns, shrimps, crabs and yabbies, and live or dead polychaete worms of the Class Polychaeta, including and not limited to bristle worms, taken from within a 10 kilometre radius of the affected farms. These may only enter or be imported into South Australia if they have been processed for human consumption as cooked product, and then securely packaged; and they are transported directly to a point of sale for human consumption".
	Outlet canal of 7ARP	Biosecurity QLD sample a single WSSV positive mud crab (qPCR CT 40.46) from the isolated water body which is the outlet canal of then WSSV negative 7ARP (For exact position see Figure 2 of the present report, or Figs. 3 and 4 in Sitrep report #9, Appendix 9). The body of water is easily accessed by the public from the road and farmers at 7ARP provide BKD with evidence it is regularly fished by members of the public.
28 Dec	Prawn farm 5IP (then 5ARP)	At 8pm BKD informed by APFA there were signs of unusual behaviour (clinical signs) in prawns from Pond 23 at 5ARP (85 days post stocking and 7 days after latest water intakes began), and that samples had been sent to BQ for testing.
29 Dec	Prawn farm 5IP	Prawns sampled from pond 23 at 5ARP on 28 <sup>th</sup> Dec confirmed to be WSSV positive by BQ (CT values range 14.0-26.0). At 9.25 am BKD rang prawn farmer who confirmed positive result and presence of clinical disease resulting in mortalities in pond 23 and problems with birds picking up dying prawns and dropping them into adjacent ponds. Bird mitigation/control officers had just arrived with scare ammunition only and succeeded in moving birds from infected ponds into other ponds on site rather than using lethal ammunition to prevent spread. Pond 23 chlorinated later that evening.
30 Dec	Prawn farm 5IP	Wind North-East 10 to 15 knots. BKD arrived at 5IP at 8.30 am to observe chlorine trucks adjacent to ponds 23 and 25. Pond 25 was emergency harvested early am that morning and was partially drained, after which remaining chlorine (approx 5 tonnes) was being placed into pond 25 as the pond level was being raised back up with water from the adjacent outlet canal. Farmers advised BKD that adjacent ponds 26, 27 and 28 had been harvested earlier (19 <sup>th</sup> Dec?, date not confirmed) with 98-100% survival. A small number of clinically diseased prawns were apparently noted by 5IP staff from pond 25 during the emergency harvest. BKD inspects surrounding ponds with BQ staff and confirms prawns with clinical signs of WSD were visible along the edges of ponds 21 and 19 with moribund prawns visible every few meters. A group of half a dozen seagulls were roosting between ponds 22 and 21 so BKD informs BQ staff that pond 21 was also likely to be infected and that bird control officers needed to look in that direction as well instead of only around pond 23. There were more birds on 5IP than at previous farms including a flock of approximately 60 seagulls between ponds 15 and 16. Bird control officers were now observed to be using a mixture of both non-lethal and lethal ammunition. A

Date	Location	Observations/comments/references
Date 30 Dec	Location	delivery of 6 new phytoplankton nets (15-20µm mesh) was delivered to the farm gate around 9.30 am and BKD worked with the on site fitter/fabricator to rig them up with poles so that plankton could be sampled from the intake canals. By 1.30 pm the next chlorine truck was already treating pond 21, however chlorine availability or equipment breakdown prevented treatment of ponds 20 and 19. All ponds on upwind (eastern) side of the northern section of the farm (ponds 1, 15, 17, 16, 18, 8 and 9) looked normal, but BKD noted one sluggish prawn on the surface along the eastern edge of pond 11 at around 4.30 pm. NE winds gusting over 15 knots in the afternoon resulted in aerosol movement of spray and foam
		being observed from pond 20 to pond 19. Plankton samples were taken from the south western edges of 5IP intake canal with the 15-20µm mesh net (including the intake trench to pond 23) late that afternoon and also after dark. Plankton were over 10 times more numerous and with more biodiversity in samples taken after dark, including several different types of copepods and insect larvae (all of which are potential WSSV vectors, see sitrep report #7, Appendix 7). Decapod crustacean activity also much increased after dark, with BKD observing large numbers of eastern king prawns on the downwind (south west) end of the intake canal after dark – visually estimated as 5-10 times more crustaceans at downwind ends of intake canals than along sides.
31 Dec	Prawn farm 5IP	Wind North-East 12 to 15 knots. BKD arrived at 5IP at 3.45 pm and advised by site controller that ponds 19, 20, 22 and 24 were chlorinated today. BKD began cast net sampling of intake canal at 7.30 pm at which time wind was North-East at 15-20 knots. Results show two to four times more prawns sampled from downwind sites and at ends of intake canals than at sites along the sides of the canal.
1 Jan 2017	Outlet canal of 7ARP	
2 Jan	Outlet canal of 7ARP	Farmer at 7ARP informs BKD that today BQ will be chlorinating the approx 80 meter x 20 meter section of outlet canal where the WSSV positive mudcrab was found.
	Prawn farm 5IP	Wind North-East 10 to 15 knots. BKD arrived at 5IP at 3.45 pm and advised by site controller that ponds 43, 44 and 45 were chlorinated today. BKD completes cast net sampling of intake canal in north-easterly wind conditions: results are >75% of prawns taken from 3 sites at ends of canals, particularly downwind. <25% of prawns taken from 3 sites upwind end of the inlet canal or along sides. Southerly change arrives with storm front and south-east wind change 15-20 knots with rain.
3 Jan	Prawn farm 7ARP	BKD advised in Technical Advisory Group phone conference that no other positive tests from mud crabs sampled in 7ARP outlet canal, nor any positive tests in production ponds at 7ARP, clearing way for them to harvest.
	Prawn farm 5IP	Wind Southerly 15 to 20 knots. BKD arrived at 5IP at 4.45 pm and advised by site controller that ponds 14, 42, 43 and 45 were chlorinated today. BKD sampled Sydney Rock Oysters ( <i>Saccostrea glomerata</i> , n = 28) at the lower intertidal zone in Logan River next to 5IP intake, dissecting and fixing gills, palps and digestive gland from 17 (min shell length 34 mm, max 98 mm, mean 61 mm shell length, SD = 15.03) in 95% ethanol and placing them in the fridge in the 5IP laboratory, and providing the remaining 11 fresh and unopened oysters to BQ for analysis. After dark BKD conducts cast net sampling of intake canal in southerly wind

Date	Location	Observations/comments/references
3 Jan		conditions. Results show >40% of prawns taken from 1 site at northern
		(downwind) end of main canal (>4 times that from previous day when
		wind was blowing from north east), confirming that distribution of
		crustaceans (and presumably their prey items) in the intake canal is
		strongly influenced by wind direction (i.e. crustaceans concentrate at the
		downwind ends of the intake canals).
4 Jan	Other tackle shops	BKD prepackages, seals and labels prawn bait samples stored in the -20°C
	in area	freezer at DigsFish and personally drives them to Eagle Farm where they
		were handed over for delivery for a retail WSSV testing program being co-
		ordinated by Matt Landos and funded by the FRDC.
5 Jan	Intake canal to 1IP	BKD informed by BQ field staff that intake canal to 1IP finally chlorinated
		in afternoon on this day.
6 Jan	Australian federal	late AM Barnaby Joyce declares via the media that imports of green prawn
	biosecurity status	products into Australia are suspended for 6 months pending investigations
		into alleged disease surveillance sample substitutions by some importers.
	Intake and outlet	Wind 5 knots from South East. BKD arrives around 2.30 pm to inspect
	canals for 7ARP	intake and outlet canals leading to/from 7ARP, interviews local worker and
		notes evidence of large amount of recreational fishing effort (discarded
		line, tackle, vehicle tracks) in several locations along intake and outlet
		canals to/from 7ARP. BKD advised that fencing and advisory signs only
		erected in the previous 2 weeks. BKD observes that these intake/outlet
		canals are narrow (7-10 meters), low flow water bodies probably
		exchanging water with Moreton Bay only on higher spring tides or when
		7ARP ponds were filled or drained, thus providing minimal dilution factor
		for any virus that may be present if WSSV positive imported prawns were
		used as bait. BKD observes (sitrep report #8, point 14, Appendix 8)
		"Indeed, it would seem prudent to fully sequence the WSSV from the
		mudcrab from the 7ARP outlet, to determine if its the same strain as the
		WSSV infecting the other farms. Given the large amount of fishing that
		historically occurred in the 7ARP drain, there may be a small chance the
		WSSV here was a separate introduction."
	Prawn farm 5IP	BKD arrives around 3 pm to change fixatives and check eradication
		progress. Biosecurity QLD data at the checkpoint reveals WSSV positive
		ponds on site at this time included 23 (index), 25, 19, 21, 7, 11, 44, 15 and
		16 (the latter two ponds being adjacent to where the flock of approximately
		60 seagulls were observed roosting by BKD on 30 December 2016).
		Ponds 4, 6, 9, 10, 11, 16 and 18 had been harvested
	Intake canal to 1IP	BKD arrives around 4.10 pm to observe intake canal to 1IP after
		chlorination. Observes advisory signage has been erected informing the
		public not to fish or remove crustaceans from that area. Biosecurity QLD
		staff were in a boat checking chlorine levels. Large numbers (several
		hundred) of dead fish and crustaceans were visible, including Greasyback
		Prawns, Jelly Prawns, Giant Shrimp (Macrobrachium sp.), and small (8-10
		cm CW) Mud Crabs, while fish included Brown Spot Estuary Cod (est. 2-3
		kg), Yellowfin Bream, Bony Bream, Silver Biddy, Sea Mullet, Glass
		Perchlets, Dusky Flathead, Freshwater Eels (est. 5-6 kg), Forktail Catfish,
		Scatophagus sp., and Butter Bream. Examination of the bank next to the
		1IP intake pipes found large numbers of live graspid crabs in their burrows
		and evidence of fishing in the form of access tracks immediately near the
		intake (Figure 10 in sitrep report #8, Appendix 8). At 5 pm BKD collects
		phytoplankton and zooplankton with 15-20µm and 100 µm plankton nets
		where the 1IP intake intersects the Logan River. With a new castnet BKD

Date	Location	Observations/comments/references
6 Jan		samples the Logan River at the 1IP intake from 5.30-6.30 pm to find mullet, numerous Yellowfin Bream, occasional Butter Bream and Glass Perchlets, as well as 15-20 small <i>Metapenaeus</i> sp., a dead mud crab and a dead prawn that probably had exited the floodgate from the chlorinated side. All of the crustaceans collected were fixed in 95% ethanol and stored on site at 1IP (Table 2). During this time conversations with farmers at 1IP disclosed that disease expression in the index pond (Pond 12) was different to other ponds that became WSSV positive at later dates. The onset of disease in the index pond 12 at 1IP was characterised by all prawns going off their feed over a day or two (Table 3). In contrast, the other ponds where birds dropped infected prawns (or other vectors spread the virus) exhibited patches of diseased prawns, but feed was still being taken by
1-7 Jan	Prawn farm 8IP (then 8ARP)	other prawns in the pond.  Farm staff inform BKD that water intakes of unknown quantity were taken into 8ARP on the top of the tide from Moreton Bay, approximate dates 1-7 <sup>th</sup> January 2017. Without water treatment this is a high risk activity as CSIRO particle tracking modelling showed that WSSV particles from upstream sites in the Logan River are likely to have been spread past this location by this date (Figure 5).
9 Jan	Prawn farm 5IP	BKD advised by BQ field staff on phone that several more ponds were now confirmed as WSSV positive on 5IP, including ponds 1 and 10, as well as 15, 16 (as noted by BKD on 6 January), bringing total number of infected ponds at 5IP to >11 (total number to be confirmed).
10 Jan	Correspondence with Biosecurity QLD CVO	BKD contacts the BQ CVO and suggests the need to conduct plankton tows in the 7ARP canal as well as in a WSSV infected pond at 5IP in order to ground truth and compare effectiveness of 15-20 vs 100 $\mu$ m (DigsFish) vs 250/500 $\mu$ m (BQ) plankton nets for detecting WSSV carriers. BKD also indicates potential for placing several species of oysters and mussels in the 7ARP canal, 5IP intake and in an infected pond at 5IP to determine their suitability as sentinels for detecting WSSV in the water. Such an experiment would help confirm if local species of shellfish can indeed concentrate WSSV, as has been recorded overseas (Vazquez-Boucard et al. 2012).
12 Jan	APFA Executive Meeting DPI Bldg	BKD attends APFA Executive meeting for presentations of data by BQ. Key data provided by BQ is presented in sitrep report #9 (Appendix 9). >2195 bait prawns tested negative for WSSV. 6 wild prawns ( <i>Metapenaeus</i> spp, <i>Acetes</i> spp.) tested positive for WSSV, all qPCR CT values >37.5. Wild Mud Crab WSSV positive qPCR CT 40.46. Only 7 positive samples so far from the wild from 6920 samples (prevalence 0.1%). Clinically diseased <i>P. monodon</i> from ponds on infected farms have qPCR CT values as low as 14. Biosecurity QLD has emergency use permit (PER83675) to use Trichlorfon (Lipidex) as a crustacide, but has not used it due to OH&S and environmental concerns.
13 Jan	Prawn farmers meeting, Yatala	Overseas expert says WSD no longer a major issue in Thailand, now they have major problems due to AHPND (Acute Hepatopancreatic Necrosis Disease) and <i>Enterocytozoon hepatopenaei</i> , both of which have emerged since 2009 and hence are not included in the Australian IRA for prawn and prawn products. WSD was largely circumvented by switching to <i>Penaeus vannamei</i> (an exotic species so not possible in Australia) stocking with SFP post larvae, filtering and treating intake water to exclude potential planktonic carriers using Trichlorfon, and minimise water exchange, and exclude birds and crabs by using crab fencing and bird netting.

Date	Location	Observations/comments/references
Date Location  13 Jan Detection recreational fisher using WSS imported prawns Logan River.		Observations/comments/references  A prawn farmer at this meeting was informed by fisheries officers whom he had met near his property that in the month or so since the area closure had been implemented for recreational fishers on the Logan River, the fisheries officers had detected 6 groups of fishers near the Alberton Boat ramp using imported raw prawns as bait. The farmer reported the officers also indicated that of the 6 bait samples confiscated and tested, 2 had returned "strong positive" results for WSSV infection. While these figures remain to be verified, the fact that recreational fishers operating near the affected prawn farms were caught using WSSV infected imported prawns for bait was officially confirmed by the Federal DAWR in a media statement that day by Deputy Secretary Lyn O'Connell as follows;  "In the course of our investigations, the department did come across recreational fishers using imported prawns labelled for human consumption for bait in the Logan River. Subsequent testing of the product did return positive results for the virus. What this tells us is that fishers using infected imported prawns for bait is one possible pathway for this disease to get into our river system and onto prawn farms and is why prawns imported for human consumption should never be used for bait." The Director of Biosecurity suspended imports of uncooked prawns earlier
		this week to ensure that pathway does not present an unacceptable risk to a currently vulnerable industry". The DAWR also stated "We are still looking at a number of pathways that may have resulted in the white spot disease incursion in Queensland, including imported feed or probiotics, contaminated equipment, or even discarded uncooked prawns - or bits of prawns - that were purchased to eat".
	Prawn farm 5IP	BKD informed by farmers that last pond at 5IP harvested today, remainder are chlorinated.
	Additional sampling sites between 4IP and 5IP	BKD identified additional environmental surveillance sampling sites in inlets between 4IP and 5IP for BQ at floodgate /bridge over sandy creek (27°42.978 S, 153°18.176 E) and a drain named Flood Structure 34 (27°43.153 S, 153°18.585 E). The BQ sampling team were contacted to see if they had sampled these locations in their investigations. They had not, but undertook to do so.
16 Jan	Meeting with Sen. Barry O'Sullivan at 5IP	DAWR staff informed Sen. O'Sullivan by phone that around 12,000 prawn consignments are received into Australia each year, and that every single batch (100%) is tested by certain NATA accredited laboratories. Sampling of each shipment consists of selecting prawns from 13 cartons selected at random. From each carton, 5 prawns are randomly selected for testing, meaning 13 x 5 = 65 prawns are sampled from each shipment, providing a theoretical 95% confidence of detecting WSSV at >5% prevalence assuming perfectly random sampling and a 100% sensitive and specific test is used. Around 41 importers select their own cartons for Government testing. Of these, 5 are under investigation for fraud and 1 is under prosecution for providing fraudulent samples (sample substitution). All 5 importers under investigation have been asked to recall their products.
20 Jan	Prawn farm 8IP (then 8ARP)	Samples of <i>P. monodon</i> taken by or given to BQ from all ponds at farm 8ARP on this date test negative for WSSV.
24 Jan	Prawn farm 8IP (then 8ARP)	Samples of <i>P. monodon</i> taken by or given to BQ from ponds 16 and 17 at farm 8ARP on this date subsequently tested as positive for WSSV (possibly subclinical infections, qPCR CT values 36-39).

Date	Location	Observations/comments/references			
25 Jan	Logan River	Trawl site #1 adjacent to 3IP, one <i>P. monodon</i> , approx 15 grams is sampled with clinical WSD (white spots on carapace). AAHL finds this prawn has a qPCR CT 14.49-16.61. An additional 9 Greasyback Prawns ( <i>Metapenaeus</i> sp.) sampled in the same trawl shot are WSSV negative.			
28 Jan	Prawn farm 8IP	BKD advised over phone by APFA that samples from 24 Jan (ponds 16, 17) confirmed as WSSV positive.			
30 Jan	Prawn farm 8IP	Biosecurity QLD informs BKD by phone that Ponds 16 and 17 have been harvested after WSSV positive detections and are now empty (ankle deep water). Only 3 ponds left unharvested, ponds 18, 19 and 20, all of which are now testing WSSV positive.			
30 Jan	Prawn farm 7IP (Then 7ARP)	Farmer informs BKD that they remain negative for WSSV despite constant testing by BQ, probably as they have not pumped water from the Logan River or Moreton Bay since early December 2016.			
2 Feb	Biosecurity QLD meeting, Yatala	Biosecurity QLD inform farmers that >3 million litres of chlorine have been used to date as part of the eradication response. Jim Thompson from BQ states in front of meeting that "The most likely pathway of introduction appears via imported prawns used as bait". The four incursion pathways being considered were:  1. WSSV enters the Logan River via infected imported prawns being used as bait/burley  2. WSSV is endemic in broodstock – vertical transmission through PLs  3. WSSV has been present in QLD waters for some time, or  4. WSSV entered in imported feed/products.  Biosecurity QLD has emergency use permit (PER83675) to use Trichlorfon (Lipidex) as a crustacide, but has not used it due to OH&S and environmental concerns (residue limit required for water discharge into the environment is <0.01µg/L) and the fact the Trichlorfon only kills the hosts and may not inactivate the virus. An APVMA permit for use of Fipronil in crab baits was also being sought.  Disposal and decontamination plans require pond chlorination (>30 mg/L >24 hrs) followed by 40 days of fallowing then re-treatment of pond water (10 mg/L chlorine >30 min) prior to discharge once chlorine levels <3 mg/L (AFANZ drinking water standard), together with crab control , followed by treatment of pond sediments with lime until day 106 then remove and dispose of upper layer of pond sediment and begin preparations for sentinel programs using P. monodon (initially planned for 14 days, now >42 days). To date all 159 of Red Fingered Marsh Crabs (Sesarma erythrodactyla, Brachyura) tested were WSSV negative (these crabs are mainly detritivores, which may explain results). First public announcement of finding of 1 P. monodon approx 15 grams sampled from trawl site #1 (Logan River opposite 3IP) with clinical WSD (white spots on carapace). AAHL finds this prawn has a qPCR CT 14.49-16.61. An additional 9 Greasyback Prawns (Metapenaeus sp.) sampled in the same trawl shot are WSSV negative. Overall prevalence in wild samples 8 positives from 8035 samples (prevalence			

Date	Location	Observations/comments/references
2 Feb		and indeed it may have been infected once it reached the Logan River, as the closest farm (3IP) had closed off its outlet canals within 2 hours of detecting WSSV on the farm (11 am on 5 Dec). However there is also a possibility that it may have been infected on nearby 1IP and escaped either during pond flushing between 25 and 30 <sup>th</sup> Nov or entered the river from the 1IP outlet canal during rainfall events prior to the 1IP outlet canal being blocked off at the river entrance and treated with hydrogen peroxide on or around 4 <sup>th</sup> Dec. Genetic testing of the prawn would be required to determine whether it was from the wild or a farm escapee.
3 Feb	Australian Federal biosecurity status	Changes to interim ban on imports of green prawn products into Australia were announced by Daryl Quinlivan, Director of Biosecurity, in a biosecurity amendment designed to lift the ban on:  (i) uncooked prawns sourced from the exclusive economic zone, or uncooked prawn meat sourced from such prawns  (ii) for uncooked prawns or uncooked prawn meat processed into dried prawns or a prawn-based food product that is shelf-stable; and  (iii) for uncooked prawns or uncooked prawn meat processed into bait for
	Prawn farm 8IP	aquatic use, pet fish food or aquaculture feed  BKD NOTES: This means that prawns exported from Australia to zones positive for WSSV (and many other crustacean diseases exotic to Australia), can again be re-imported without the need for further testing, with inherent risks of cross contamination and/or product substitution for same or different species of prawns from other countries. The risk profile for this activity is not clearly articulated by the department, especially considering the recent (post-2009) publication of new phytosanitary information on risks of viable WSSV and other new (emergence post-2009) diseases being transmitted via various commodities (see discussion below and papers by Overstreet et al. (2009), Ma et al. (2009), Stentiford et al. (2009), Oidtmann and Stentiford (2011), Reddy et al. (2011a, 2011b), Bateman et al. (2012), Stentiford et al. (2012), Stentiford (2012), Jones (2012), Shields (2012), Behringer (2012), Lightner et al. (2012), Tran et al. (2013a, 2013b), Reddy et al. (2013), Nunan et al. (2014), De La Pena et al. (2015), Cowley et al. (2015), Thitamadee et al. (2016), amongst many others), thus these activities should be subjected to a full review as part of a full update of the now well outdated 2009 prawn Import Risk Analysis.  Wind 10 knots from North. BKD arrives around 11 am to inspect ponds
	Prawn farm 81P	Wind 10 knots from North. BRD arrives around 11 am to inspect ponds 16-20 at 8IP and take plankton and invertebrate samples from WSSV positive ponds 18, 19, and 20. BKD was denied access to the farm for sampling on the afternoon of 2 Feb, as were BQ staff on several prior occasions. Today access was allowed, but no bird control officers were observed on site. Farm staff advised BKD the farm has its own hatchery and stocked its own ponds, and had not taken in water since the first week in January (around 3 weeks). En-route to pond 18 BKD observes several hundred birds wading in ankle deep water in ponds 16 and 17 (Figures 6, 7). All of the ponds on this farm were either already harvested (up to pond 17) or in the final processes of drain harvesting (ponds 18, 19, 20) with very little water remaining in most ponds or in the settling ponds, however the intake canal appeared nearly full. Dozens of live, unharvested <i>P. monodon</i> were observed moving around in the shallow water in ponds 18, 19, and 20, while a small number (<5) of dead prawns were observed in ponds 18 and 20. Samples from plankton tows (15-20μm mesh and 100μm

Date	Location	Observations/comments/references
3 Feb		mesh) were taken from each of the ponds 18, 19 and 20 and fixed in 70% ethanol. Samples of live barnacles were also fixed in ethanol from ropes holding equipment on the bottom of pond 18, while a single recently deceased <i>P. monodon</i> from pond 20 was also fixed in ethanol. All samples
		were provided to BQ staff who visited the farm that afternoon (Table 2). BKD left the plankton nets on site, and decontaminated boots hands and equipment prior to leaving the site. BKD also informs BQ staff of the risk of disease spread posed by birds wading in WSSV infected ponds both verbally and via text message sent 4.10 pm on 3 Feb 2017.
5 Feb	Prawn farm 8IP	Biosecurity QLD advises BKD that chlorination was started today in all ponds (16, 17, 18, 19, 20) at 8IP.
8 Feb	Trawl sample site #8 opposite 8IP	Announcement by BQ that around 100 <i>P. monodon</i> sampled from Trawl site #8 test positive for WSSV. Prevalence data is not available as samples were batched (20 batches of 5 prawns) prior to testing at EAMI. BKD NOTES: Most likely these prawns are escapees from farms in the area, as natural <i>P. monodon</i> stocks are rare in Moreton Bay, however genetic testing may be needed to definitively reveal their origins. Prawn post larvae are stocked at small sizes and can sometimes escape around monk boards or during water releases. However, this does not mean these prawns were infected on a farm then escaped, and indeed they may have been infected once they reached southern Moreton Bay. However, its not clear at the time of publication whether the closest farm (8IP) had all its outlet canals properly blocked off. If the outlet canals were not completely
10 Feb	Imported Prawn Suspension Update	blocked, there is also a possibility that the prawns may have been infected on nearby 8IP and escaped during pond drain harvesting activities.  The DAWR "Imported Prawn Suspension Update" for 10 Feb 2017 said: The following product categories are still being considered as part of the
		review:  o Australian sourced prawns sent overseas for processing o mixed seafood consignments containing uncooked prawns o marinated prawns.  * It may take up to 8 weeks to review import conditions for these products. This timeframe is calculated from the initial date of suspension.  * Once these products have been reviewed, a decision can be made on possible future risk management options to allow for the safe resumption of trade.
		* The Department of Agriculture and Water Resources is accepting submissions from parties with proposals about how they believe their particular situation manages the biosecurity risks associated with import of uncooked prawns or prawn products which are currently suspended under the Biosecurity (Suspended Good - Uncooked Prawns) Determination 2017.  * The department will conduct a risk assessment of the proposal and using the outcomes of that risk assessment, the Director of Biosecurity will make a decision about whether the product can be exempt from the temporary suspension, or if it must remain suspended from import until further biosecurity risk assessments can be completed.  Enhanced measures for goods in transit  * The department is currently working through over 100 consignments of
		prawns which were already in transit to Australia when the suspension took effect.

Date	Location	Observations/comments/references
		* These consignments are being subjected to an enhanced inspection and
10 Feb		double testing regime at the border to ensure any product entering the
		country is free from white spot disease. The product will not be released
		until it has undergone double testing and is confirmed free from the
		disease.
		Number of batches fully tested 37
		Number of batches released 11
		Number of batches refused 26
		BKD NOTES: These latest data represent a rejection rate at the international border of 70.3% (26/37) using proper testing procedures. This is higher than the 66% prevalence detected by researchers in the EU who surveyed imported green <i>P. vannamei</i> prawns at British supermarkets
		(Bateman et al. 2012). The 70.3% rejection rate at the border under proper surveillance needs to be compared to the 16.3% rejection rate (73/448 consignments) reported by DAWR in the phone conference of 15 Dec 2016 (see above) for shipments between May and December 2016. The
		difference in these two figures $(70.3 - 16.3)$ is 54%, which represents the likely prevalence of WSSV infected prawns that cleared quarantine in
		Australia and would be expected to be available to consumers at retail counters in the period leading up to Christmas/New Year 2016.
	Discharge of an	OIE followup report #10. Laboratory qPCR tests confirmed WSSV DNA
	infected premise	in one Mud Crab (Scylla serrata) sampled from the discharge channel of an
	infected prefinse	infected premise.
11 Feb	Prawn farm 7IP	Farmer informs BKD suspect clinical WSD observed in <i>P. monodon</i>
	(Then 7ARP)	examined from Pond 4 at 7ARP. Samples sent to BQ for testing.
	Meeting with Sen.	BKD meets with Sen. Ruston and helps show her around infected farm
	Anne Ruston	sites and attend meetings with farmers.
13 Feb	Prawn farm 7IP	Farmer informs BKD that WSSV confirmed in <i>P. monodon</i> sampled from
		pond 4 at (now) 7IP. Given the fact that 7IP has not pumped any water
		from the Logan River or Moreton Bay since early December 2016 (2
		months), and the fact that many hundreds of bird were observed by BKD
		wading in WSSV infected ponds 16 and 17 at 8IP (around 600-750 meters
		to the east) on 3 Feb 2017, it appears temporally and spatially plausible that
		the introduction of WSSV into pond 4 at 7IP was via movement of birds
		between 8IP and 7IP, especially as the birds from ponds 16 and 17 would
		have been disturbed when those ponds were chlorinated (c. 5 Feb 2017).
14 Feb	Prawn farm 7IP	Farmer advises media that chlorination of production ponds has
		commenced at 7IP.
16 Feb	Australian Federal	Senate Inquiry into WSSV disease incursion announced
	biosecurity status	



Figure 3. Moribund Black Tiger Prawns (*Penaeus monodon*) sampled from pond 11 at 1IP on 2 December 2016, displaying clinical signs of WSD. The range of pondside presentations is shown in the lower picture.

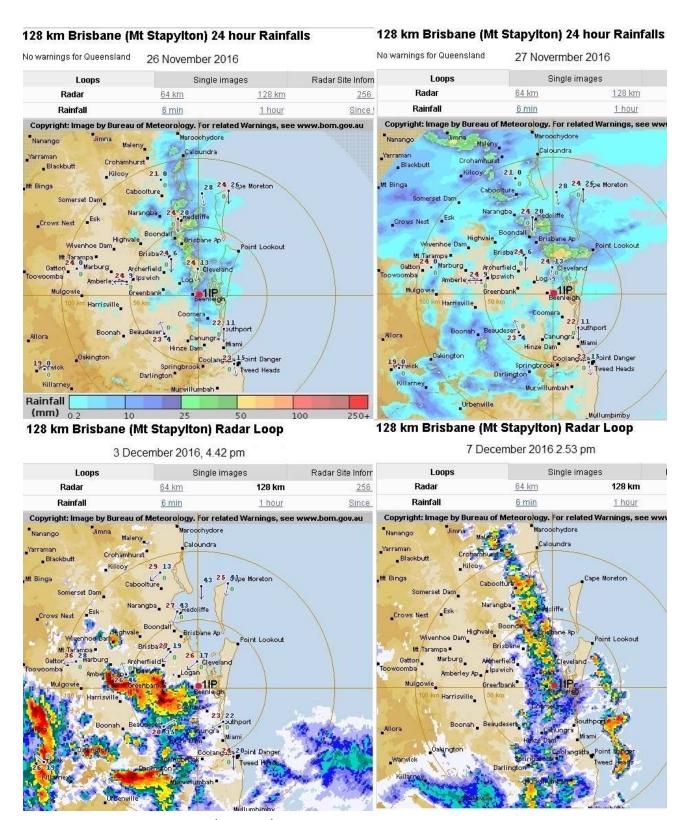


Figure 4. Rainfall data for 26<sup>th</sup> and 27<sup>th</sup> November 2016 (top) and 3 and 7 December 2016 (bottom). Moderate rain events (57 mm at Logan River Water Treatment Plant 26<sup>th</sup> Nov, storms on 3<sup>rd</sup> Dec) suggest runoff into 1IP outlet canal would occur, increasing risk of water from initial pond flushing reaching the Logan River prior to blocking of the 1IP canal at the river end sometime around 4<sup>th</sup> December.

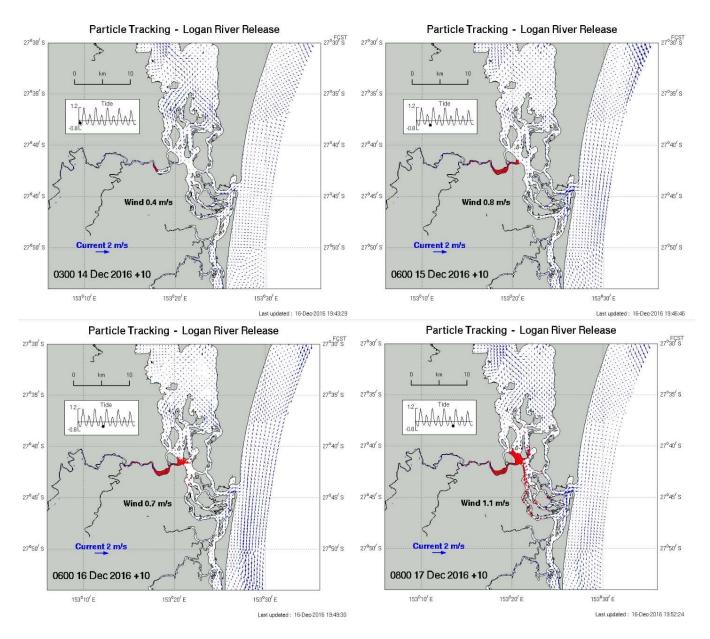


Figure 5. CSIRO particle tracking modelling for movements of water particles (red dots) from the middle reaches of the Logan River near farms 1IP, 2IP and 3IP towards the mouth of the Logan River at low tide from 14-17 December 2016 using basic assumptions of 2 meters/sec seawards current flow in the river. Note: these simulations were preliminary and indicative only, and may not represent actual particle movements.



Figure 6. Hundreds of birds were evident wading through WSSV positive ponds 16 and 17 (above) at 8IP on 3<sup>rd</sup> February 2017 at around 11.30 am. These ponds were drain harvested at that time, but not chlorinated /decontaminated until Sunday 5<sup>th</sup> February. In the absence of bird control officers on 8IP at the time, these birds could act as vectors allowing movement of WSSV to nearby farms, including (then) 7ARP around 600-750 meters to the west of these ponds, which experienced WSD 11<sup>th</sup> Feb, 8 days after this photo was taken.



Figure 7. Closeup of birds in pond 17 at 8IP on 3<sup>rd</sup> February 2017. Several species of wading birds were noted including egrets, pelicans and ibis.

## 3.2 Additional data from field sampling

Table 2. Samples collected by BKD during field observations of an outbreak of WSD in prawn farms on the Logan River.

Date	Location sampled	Sample type	Storage	
				location
2 Dec	1IP (pond 11)	Cultured prawns ( $P. monodon$ ), $n = 5$	Davidsons fixative	1IP
2016				
5 Dec	3IP (pond 2)	Cultured prawns ( $P. monodon$ ), $n = 2$	Davidsons fixative	3IP
		Red fingered crabs $n = 2$	95% ethanol	3IP
11 Dec	Tackle shop (9IP)	Wild banana prawns, Metapenaeus spp., F.	95% ethanol, or	Siezed by
		Paleomonidae n = approx 20	frozen	Biosecurity
				QLD
11 Dec	3IP pond 1	Cultured prawns ( $P. monodon$ ), $n = 60$	95% ethanol	3IP
	3IP pond 5	Cultured prawns ( $P. monodon$ ), $n = 60$	95% ethanol	3IP
12 Dec	3IP pond 6	Cultured prawns ( $P. monodon$ ), $n = 60$	95% ethanol	3IP
	3IP pond 7	Cultured prawns ( $P. monodon$ ), $n = 60$	95% ethanol	3IP
12 Dec	4IP pond 18	Cultured prawns ( $P. monodon$ ), $n = 60$	95% ethanol	4IP
13 Dec	2IP pond 22	Cultured prawns ( $P. monodon$ ), $n = 60$	95% ethanol	2IP
13 Dec	4IP pond 10	Cultured prawns ( <i>P. monodon</i> ), n = 60	95% ethanol	4IP
	4IP pond 11	Cultured prawns ( $P. monodon$ ), $n = 60$	95% ethanol	4IP
	4IP pond 17	Cultured prawns ( $P. monodon$ ), $n = 60$	4IP	
15 Dec	2IP pond 17	Cultured prawns ( <i>P. monodon</i> ), n = 60	2IP	
30 Dec	5IP intake	Plankton tows $20\mu m$ net $n = 6$	95% ethanol	5IP
	5IP intake	Eastern king prawns $n = 12$	95% ethanol	5IP
31Dec	5IP intake	Penaeus spp, Metapenaeus spp. n = 60	95% ethanol	5IP
2 Jan	5IP intake	Penaeus spp, Metapenaeus spp. n = 14	95% ethanol	Supplied to
2017		Penaeus spp, Metapenaeus spp. n = 14	Live chilled	Biosecurity
				QLD staff
3 Jan	Logan river near 5IP Sydney rock oysters n = 17 95% ethanol		5IP	
	intake pumps	Sydney rock oysters $n = 11$	Live	Supplied to
				Biosecurity
			95% ethanol	QLD staff
3 Jan	5IP intake	Penaeus spp, Metapenaeus spp. n = 120	5IP	
6 Jan	1IP intake near river			1IP
		Metapenaeus spp. n =20, Scylla sp. n=1	95% ethanol	1IP
13 Jan	7ARP outlet	Plankton tow 20 and $100\mu m$ net $n = 1$ ea	7ARP	
3 Feb	8IP pond 18	Plankton tow 20 and $100\mu m$ net $n = 1$ ea	95% ethanol	Supplied to
	8IP pond 18	P pond 18 Live barnacles n = approx 30 95%		Biosecurity
	8IP pond 19	Plankton tow 20 and $100\mu m$ net $n = 1$ ea	95% ethanol	QLD staff
	8IP pond 20	Plankton tow 20 and $100\mu m$ net $n = 1$ ea	95% ethanol	
	8IP pond 20	Recently dead <i>P. monodon</i> n = 1	95% ethanol	

Table 3. Water quality data for index pond 12 at 1IP in the 6 weeks leading up to the index WSD outbreak. (dps = days post stocking)

Date	Water	Salinity	рН	Dissolved	Event notes
	Temp (°C)	(ppt)		Oxygen (mg/L)	
15 Sept 2016					Pond stocked
10 Oct 2016	20.8	16.6	8.06	8.17	
11 Oct	21.9	16.81	7.97	6.83	
12 Oct	20.3	16.95	7.91	7.00	
13 Oct	20.4	17.13	7.79	6.97	
17 Oct	19.9	17.89	7.98	6.91	
19 Oct	20.3	18.47	7.79	6.53	
20 Oct	20.7	18.88	7.77	7.74	
21 Oct	21.5	19.36	7.77	8.29	
24 Oct	19.8	19.91	7.73	7.31	
25 Oct	20.0	20.38	7.73	7.45	
26 Oct	21.0	20.57	7.57	7.17	
27 Oct	20.9	21.02	7.63	5.43	
28 Oct	22.2	21.48	7.51	7.60	
31 Oct	22.4	22.03	7.35	5.81	
1 Nov 2016	22.7	22.20	7.34	6.04	
2 Nov	22.4	22.60	7.50	6.71	
3 Nov	22.2	23.00	7.52	6.99	
4 Nov	22.3	23.41	7.54	6.37	
10 Nov	24.3	24.71	7.33	4.89	
11 Nov	24.6	24.83	7.34	5.86	
14 Nov, 60 dps	23.8	26.83	8.14	7.26	Water intake (pH and DO increases)
15 Nov	22.1	27.37	8.07	6.08	
16 Nov, 62dps	21.9	27.71	7.78	6.35	Drop in water temperature
17 Nov	22.6	28.09	7.85	6.17	
18 Nov	21.9	28.13	7.95	6.12	
21 Nov	23.3	28.58	7.76	5.29	
22 Nov, 68 dps	23.3	28.76	7.61	5.47	Prawns off feed, first clinical signs
23 Nov, 69 dps	23.7	29.63	7.44	5.7	First WSD mortalities observed
24 Nov	Data collection discontinued due to disease (to reduce risk of disease spread via equipment)				

### 3.3 DAWR Reporting to OIE

A very useful source of information is the reports provided to the OIE by the Office of the Chief Veterinarian in the Federal Department of Agriculture and Water Resources (DAWR). These are summarised below in Table 4, followed by a summary of the main points from each report.

Table 4. Summary of reports from Australia to the OIE during this WSD outbreak up until present date of publication.

Date	Report	Report Link
submitted	Type	
to OIE		
1 Dec	Immediate	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=21737
2016	Notification	
8 Dec	Follow-up	http://www.oie.int/wahis 2/public/wahid.php/Reviewreport/Review?reportid=21830
2016	Report 1	
15 Dec	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=21926
2016	Report 2	
22 Dec	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=21987
2016	Report 3	
29 Dec	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=22126
2016	Report 4	
12 Jan	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=22277
2017	Report 5	
19 Jan	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=22416
2017	Report 6	
25 Jan	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=22553
2017	Report 7	
1 Feb	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=22645
2017	Report 8	
8 Feb	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=22770
2017	Report 9	
16 Feb	Follow-up	http://www.oie.int/wahis_2/public/wahid.php/Reviewreport/Review?reportid=22856
2017	Report 10	

Content of reports from Australia to the OIE during this WSD outbreak up until date of publication.

#### **Immediate Notification (1/12/2016)**

http://www.oie.int/wahis 2/public/wahid.php/Reviewreport/Review?reportid=21737

**Report Type:** Immediate Notification **Date of start of event:** 22/11/2016 **Date of confirmation of event:** 1/1/2016 **Report date:** 1/12/2016

**Date submitted to OIE:** 1/12/2016 **Reason for Notification:** First occurrence of a listed

disease

Manifestation of disease: Clinical disease Causal agent: White spot syndrome virus

**Nature of diagnosis:** Laboratory (advanced) **This event pertains to:** the whole country

New Outbreaks: (1)

Outbreak 1: Logan River, Oueensland

Date of start of outbreak:5/12/2016Status:Resolved (10/12/2016)Epidemiological unit:PondWater Type:Salt WaterPopulation Type:FarmedProduction system:Semi-closed

**Affected animals:** Giant tiger prawn (*P. monodon*) **Affected population:** Pond reared *P. monodon* 

Source of the outbreak(s) or origin of infection: Unknown or inconclusive

#### **Epidemiological comments**

On Tuesday 22 November 2016, a prawn farmer reported a minor mortality event with a small numbers of dead prawns, reduced feeding and some unusual swimming behaviour. Samples were collected on 22 and 25 November 2016 and processed with routine priority. Higher mortalities were reported from one pond on

28 November 2016. Positive PCR results from Queensland's Biosecurity Sciences Laboratory were reported on 30 November 2016 and confirmed by the Australian Animal Health Laboratory on 1 December 2016. Measures to contain the disease to the affected farm have been applied. The disease response aims to eradicate the disease from the affected farm and confirm freedom in nearby farms and in wild crustaceans. The source of the outbreak is unknown.

**Diagnostic Test Results** 

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** gene sequencing **Test date:** 1/12/2016 **Result:** Positive

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** polymerase chain reaction (PCR) **Test date:** 1/12/2016 **Result:** Positive

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 1/12/2016 **Result:** Positive

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 1/12/2016 **Result:** Positive

**Laboratory:** QLD Biosecurity Sciences Laboratory Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** histopathological examination **Test date:** 30/11/2016 **Result:** Positive

**Laboratory:** QLD Biosecurity Sciences Laboratory Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** polymerase chain reaction (PCR) **Test date:** 30/11/2016 **Result:** Positive

**Control Measures** 

**Measures applied:** Traceability, Disinfection/Disinfestation, Quarantine, Surveillance outside containment and/or protection zone, Stamping out, Official disposal of carcasses, by-products and waste, Surveillance within containment and/or protection zone, Control of wildlife reservoirs, zoning, Vaccination permitted (if a vaccine exists), no treatment of affected animals.

### Follow-up Report 1 (8/12/2016)

http://www.oie.int/wahis 2/public/wahid.php/Reviewreport/Review?reportid=21830

**Report Type:** Follow-up Report No. 1

New Outbreaks: (2)

Outbreak 1: Logan River, Oueensland

Date of start of outbreak:5/12/2016Status:Resolved (16/12/2016)Epidemiological unit:PondWater Type:Salt WaterPopulation Type:FarmedProduction system:Semi-closed

Formation Type: Farmed Froduction system: Semi-closed

**Affected animals:** Giant tiger prawn (*P. monodon*) **Affected population:** Pond reared *P. monodon* 

Outbreak 2: Logan River, Queensland

**Date of start of outbreak:** 8/12/2016 **Status:** Continuing (or date resolved not provided)

**Epidemiological unit:** River system **Water Type:** Salt Water **Population Type:** Wild **Production system:** Open

**Affected animals:** Prawns (*Penaeus* spp.) **Affected population:** wild prawns collected from Logan

River

**Source of the outbreak(s) or origin of infection:** Unknown or inconclusive

**Epidemiological comments** 

Measures to contain the disease to the index farm have been applied. Destruction of stock and decontamination of all ponds and drainage channels on this farm is near completion. Tracing identified two nearby farms that were epidemiologically linked to the index farm. Clinical disease was observed on one of these farms on 5 December 2016 and containment measures applied. Destruction and decontamination has commenced. Surveillance of six other farms in the region, and from wild populations of crustaceans in the Logan River is ongoing. On 8 December 2016, some prawns sampled from a section of the Logan River tested PCR positive. Further sampling will be undertaken to determine whether disease has established in the environment.

**Diagnostic Test Results** 

**Laboratory:** AAHL **Species:** Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 8/12/2016 **Result:** Positive

#### Follow-up Report 2 (15/12/2016)

http://www.oie.int/wahis\_2/public/wahid.php/Reviewreport/Review?reportid=21926

**Report Type:** Follow-up Report No. 2

New Outbreaks: (2)

Outbreak 1: Logan River, Queensland

Date of start of outbreak:8/12/2016Status:Resolved (16/12/2016)Epidemiological unit:PondWater Type:Salt WaterPopulation Type:FarmedProduction system:Semi-closed

**Affected animals:** Giant tiger prawn (*P. monodon*) **Affected population:** Pond reared *P. monodon* 

Outbreak 2: Logan River, Queensland

Date of start of outbreak:12/12/2016 Status:Resolved (21/12/2016)Epidemiological unit:PondWater Type:Salt WaterPopulation Type:FarmedProduction system:Semi-closed

**Affected animals:** Giant tiger prawn (*P. monodon*) **Affected population:** Pond reared *P. monodon* 

Source of the outbreak(s) or origin of infection: Unknown or inconclusive

**Epidemiological comments** 

Destruction of stock and decontamination of all ponds and drainage channels on the index farm and the farm identified on 5 December is complete. Symptoms on two nearby farms were observed on 8 December and 12 December. Affected ponds were immediately treated and destruction and decontamination of all ponds and channels on both farms is underway. Daily surveillance of 4 other farms in the region, and from wild populations of crustaceans in the Logan River is ongoing. Since 8 December over 1200 samples of prawns and crabs from the Logan River have been PCR negative for the virus. Further sampling is being undertaken to provide early detection of new disease outbreaks in farms and determine whether disease has established in the environment.

### **Diagnostic Test Results**

**Laboratory:** AAHL **Species:** Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 9/12/2016 **Result:** Positive

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 15/12/2016 **Result:** Positive

**Laboratory:** QLD Biosecurity Sciences Laboratory **Species:** Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 13/12/2016 **Result:** Positive

#### Follow-up Report 3 (22/12/2016)

http://www.oie.int/wahis 2/public/wahid.php/Reviewreport/Review?reportid=21987

**Report Type:** Follow-up Report No. 3

**Outbreaks:** There are no new outbreaks in this report

**Source of the outbreak(s) or origin of infection:** Unknown or inconclusive

**Epidemiological comments** 

Destruction of all stock on all four infected premises is complete. All ponds and channels have been chlorinated. A disposal and decontamination plan is being implemented. All other prawn farms in the area are under intensive daily visual surveillance for behavioural changes, and regular sampling and testing by PCR. Decapod samples from the river and surrounding areas are being collected daily and tested. To date over 5000 samples have been tested with no confirmed positive results from wild caught samples since 8 December.

#### Follow-up Report 4 (29/12/2016)

http://www.oie.int/wahis\_2/public/wahid.php/Reviewreport/Review?reportid=22126

**Report Type:** Follow-up Report No. 4

New Outbreaks: (1)

Outbreak 1: Logan River, Queensland

**Date of start of outbreak:** 28/12/2016 **Status:** Continuing (or date resolved not provided)

**Epidemiological unit:** Pond **Water Type:** Salt Water **Population Type:** Farmed **Production system:** Semi-closed

**Affected animals:** Giant tiger prawn (*P. monodon*) **Affected population:** Pond reared *P. monodon* 

**Source of the outbreak(s) or origin of infection:** Unknown or inconclusive

**Epidemiological comments** 

Destruction of all stock on four infected premises is complete. All ponds, settlement ponds and channels have been treated. A disposal and decontamination plan is being implemented. All other prawn farms in the area are under intensive daily visual surveillance for behavioural changes, and regular sampling and testing by PCR. One of these farms had confirmed PCR positive results for WSSV from one pond on 29 December. This pond was treated and stock destroyed on 29 December. Increased surveillance measures are in place. Decapod samples from the river and surrounding areas are being collected regularly and tested. There have been no confirmed positive results from wild caught samples since 8 December.

**Diagnostic Test Results** 

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 29/12/2016 **Result:** Positive

### Follow-up Report 5 (12/1/2017)

http://www.oie.int/wahis\_2/public/wahid.php/Reviewreport/Review?reportid=22277

**Report Type:** Follow-up Report No. 5

**Outbreaks:** There are no new outbreaks in this report

**Source of the outbreak(s) or origin of infection:** Unknown or inconclusive

**Epidemiological comments** 

Destruction of all stock on four infected premises is complete. All ponds, settlement ponds and channels have been treated. A disposal and decontamination plan is being implemented. The fifth infected farm is undergoing treatment by chlorination, and emergency harvest of uninfected ponds. The other prawn farms in the area are under intensive daily visual surveillance for behavioural changes, and regular sampling and testing by PCR. Three of 15 crab samples (including *Scylla serrata*) taken from channels adjacent to the current infected farm have been confirmed positive for WSSV by PCR. Channels will be treated as part of the treatment plan for this premises. One PCR positive crab has been sampled on another farm channel. The channel has been treated and no further positive detections have been made in that location. Decapod samples from the river and surrounding areas are being collected regularly and tested. There have been no confirmed positive results from samples from the river since 8 December. Samples of wild decapods in the area (estuaries and coastal waters north or east of the outbreak zone) have also tested negative by PCR. Additional sampling of potential hosts (including oysters, copepods and polychaetes) is underway. Testing by the regional laboratories and the national laboratory is ongoing on a daily basis.

**Diagnostic Test Results** 

**Laboratory:** AAHL **Species:** Crabs (Decapoda)

**Test:** Real time PCR **Test date:** 10/01/2017 **Result:** Positive

#### Follow-up Report 6 (19/1/2017)

http://www.oie.int/wahis\_2/public/wahid.php/Reviewreport/Review?reportid=22416

**Report Type:** Follow-up Report No. 6

**Outbreaks:** There are no new outbreaks in this report

Source of the outbreak(s) or origin of infection: Unknown or inconclusive

**Epidemiological comments** 

Destruction of all stock on four infected premises is complete. All ponds, settlement ponds and channels have been treated. A disposal and decontamination plan is being implemented. Production ponds on the fifth infected farm have all been treated by chlorination. Two mud crabs (*Scylla serrata*) sampled from channels on the infected farm before completion of treatment were found to be qPCR positive for WSSV. The other prawn farms in the area are under intensive daily visual surveillance for behavioural changes, regular sampling and testing by PCR, and are progressively early harvesting stocks. Decapod samples from the river and surrounding areas are being collected regularly and tested. There have been no confirmed positive results from samples from the river since 8 December. Samples of wild decapods in the area

(estuaries and coastal waters north or east of the outbreak zone) have also tested negative by PCR. Over 8000 samples from wild populations have been tested to date. Additional sampling of potential hosts (including oysters, copepods and polychaetes) is underway. Testing by the regional laboratories and the national laboratory is ongoing on a daily basis.

**Diagnostic Test Results** 

**Laboratory:** AAHL **Species:** Crabs (Decapoda)

**Test:** Real time PCR **Test date:** 18/01/2017 **Result:** Positive

### Follow-up Report 7 (25/1/2017)

http://www.oie.int/wahis\_2/public/wahid.php/Reviewreport/Review?reportid=22553

**Report Type:** Follow-up Report No. 7

**Outbreaks:** There are no new outbreaks in this report

**Source of the outbreak(s) or origin of infection:** Unknown or inconclusive

### **Epidemiological comments**

Destruction of stock on all five infected premises is complete. All production ponds and associated water bodies on four of the infected premises have been treated. Some infrastructure (channels and settlement ponds) will be treated on the fifth infected premises this week. A disposal and decontamination plan is being implemented. Laboratory qPCR tests detected WSSV DNA in one mud crab (*Scylla serrata*) sampled from an outlet channel on the fifth infected farm before completion of treatment. The other prawn farms in the area are under intensive daily visual surveillance for behavioural changes, regular sampling and testing by PCR, and are progressively early harvesting stocks. Decapod samples from the river and surrounding areas are being collected regularly and tested. There has been no detection of WSSV DNA from samples from the river since 8 December. Over 8000 samples from wild populations have been tested to date. Additional sampling of potential hosts (including oysters, copepods and polychaetes) is underway. Initial PCR testing of oysters from the river has not detected WSSV DNA. Testing by the regional laboratories and the national laboratory is ongoing on a daily basis.

**Diagnostic Test Results** 

**Laboratory:** AAHL **Species:** Crabs (Decapoda)

**Test:** Real time PCR **Test date:** 25/01/2017 **Result:** Positive

### **Follow-up Report 8 (1/2/2017)**

http://www.oie.int/wahis\_2/public/wahid.php/Reviewreport/Review?reportid=22645

**Report Type:** Follow-up Report No. 8

**Outbreaks:** There are no new outbreaks in this report

**Source of the outbreak(s) or origin of infection:** Unknown or inconclusive

#### **Epidemiological comments**

Destruction of stock on all five infected premises is complete. All production ponds and associated water bodies on the infected premises have been treated. A disposal and decontamination plan is being implemented to dispose of water and sediments from ponds. Of the two at risk premises in the area, one has completed harvest of stock. The other prawn farm still has stock and is under intensive daily visual surveillance for behavioural changes, regular sampling and testing by PCR, and is progressively harvesting. Decapod samples from the river and surrounding areas are being collected regularly and tested. Laboratory qPCR tests detected WSSV DNA in one giant tiger prawn (*Penaeus monodon*) sampled from the Logan River on 27 January. With the exception of this new result, there has been no detection of WSSV DNA from samples from the river since 8 December. Over 10,000 samples from wild populations have been tested to date. Additional sampling of potential hosts is underway. Testing by the regional laboratories and the national laboratory is ongoing on a daily basis.

**Diagnostic Test Results** 

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 31/01/2017 **Result:** Positive

### **Follow-up Report 9 (8/2/2017)**

http://www.oie.int/wahis 2/public/wahid.php/Reviewreport/Review?reportid=22770

**Report Type:** Follow-up Report No. 9

**Outbreaks:** There are no new outbreaks in this report

**Source of the outbreak(s) or origin of infection:** Unknown or inconclusive

**Epidemiological comments** 

Destruction of stock on all infected premises is complete. A disposal and decontamination plan is being implemented to dispose of water and sediments from ponds, and decontaminate water bodies on farms including channels and settlement ponds. The remaining uninfected prawn farm still has stock and is under intensive daily visual surveillance for behavioural changes, regular sampling and testing by PCR, and is progressively harvesting. Decapod samples from the river and surrounding areas are being collected regularly and tested. On February 4 laboratory qPCR tests detected WSSV DNA in giant tiger prawns (*Penaeus monodon*) sampled from a location adjacent to the river mouth outside the most recently infected premises. Additional sampling of potential hosts is ongoing. Testing by the regional laboratories and the national laboratory is ongoing on a daily basis. The response to WSD is still aimed at eradication.

**Diagnostic Test Results** 

**Laboratory:** AAHL Species: Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 4/02/2017 **Result:** Positive

### Follow-up Report10 (16/2/2017)

http://www.oie.int/wahis 2/public/wahid.php/Reviewreport/Review?reportid=22856

**Report Type:** Follow-up Report No. 10

New Outbreaks: (1)

Outbreak 1: Logan River, Queensland

Date of start of outbreak:11/02/2017Status:Resolved (14/2/2017)Epidemiological unit:PondWater Type:Brackish WaterPopulation Type:FarmedProduction system:Semi-closed

**Affected animals:** Giant tiger prawn (*P. monodon*) **Affected population:** Pond reared *P. monodon* 

Source of the outbreak(s) or origin of infection: Unknown or inconclusive

**Epidemiological comments** 

The remaining uninfected prawn farm in the Logan River area detected signs of diseased prawns on 11 February and presence of WSSV DNA was confirmed on 13 February. The infected pond was treated on 11 February and remaining ponds were destocked and are being treated. Destruction of stock on all farms in this area is complete, and treatment of all production facilities will be completed soon. A disposal and decontamination plan is being implemented to dispose of water and sediments from ponds in a biosecure manner. Decapod samples from the river and surrounding areas are being collected regularly and tested. On 10 February laboratory qPCR tests confirmed WSSV DNA in one mud crab (*Scylla serrata*) sampled from the discharge channel of an infected premises. Additional sampling of potential hosts is ongoing. Testing of samples collected under the surveillance program is ongoing on a daily basis by regional laboratories and the national laboratory. The response objective is eradication.

**Diagnostic Test Results** 

**Laboratory:** AAHL **Species:** Crabs (Decapoda)

**Test:** Real time PCR **Test date:** 10/02/2017 **Result:** Positive

**Laboratory:** AAHL **Species:** Giant tiger prawn (*Penaeus monodon*)

**Test:** Real time PCR **Test date:** 13/02/2017 **Result:** Positive

# 4.0 Discussion and Recommendations

At the time of publication of this document, the eradication response by BQ was still underway with the objective of regaining freedom from WSD in the Logan River area. However the incursion has already provided many poignant lessons regarding the substantial biological, economic and human consequences associated with a biosecurity breakdown of this magnitude. The nearest region where WSSV is recognized as endemic is probably somewhere in eastern Indonesia, hence this incursion represents a significant shift in the distribution of WSSV in the Oceania region. Because of this, there are many unknowns regarding the behaviour of WSSV in this new environment, so the ultimate outcome of the incursion cannot be forseen at this time. Indeed, at the time of writing it is expected that it will probably be sometime in late 2017 before we have any understanding of whether this unwanted pathogen will fizzle out, or become established in the Logan River and/or southern Moreton Bay.

### 4.1 Recommendations relating to the response

Even though it is still early days, and too soon to discuss the effectiveness of decontamination and disposal efforts, some hard earned lessons have already been learned. In relation to the response to the disease outbreak itself, these include:

- It is important to consider the potential for an exotic disease incursion in any differential diagnosis list (mentioned in relation to advice to farmers allowing options for pond flushing). In view of the situation at the international border with quarantine breakdowns involving uncooked prawn commodities, preparedness and heightened surveillance for exotic diseases could have been facilitated if Federal authorities had communicated the increased risk to state authorities.
- Decisions made at the earliest stages of an incursion response may have significant impacts on the ultimate outcomes and chances of eventual eradication success. A rule of thumb may be (unless proven otherwise) to imagine or assume the worst case scenario and attempt to cater for it, while hoping for the best. (mentioned in relation to the decision to block 1IP outlet canal at the exit of the settlement pond instead of where it enters the Logan River).
- Restricting activities of people and movements of animals in the control zone surrounding affected farms is important (mentioned in relation to unrestricted movements of recreational fishers in the inlet canals for at least one week after WSSV was known to be present, and failure to erect signage advising no movements of crustaceans for more than 3 weeks). Enforcement is necessary and useful for preventing movements of potentially infected animals and materials, as well as for gathering information, as shown by the subsequent detections of recreational fishers using WSSV positive imported prawns as bait near the infected farms.
- Availability of biosecurity field staff with specialist aquatic animal training is limited. Water bodies
  which hide the animals within them can be very hard to "read" without some sort of specialist
  training. Lack of training for personnel on the ground may hinder information transfer to decision
  makers meaning important decisions may have to be made by people who are remote from the

situation on the ground and/or who may not have even visited the area where the incident/incursion is occurring. Having more aquatic trained decision makers on the ground on farms and scanning the wider area for potential biosecurity leaks (e.g. local bait from the river being sold in tackle shops in this case) would have been advantageous and would have allowed more precise decision making and more rapid adaptation to changing situations.

- Similarly, from the coalface the response structure appeared unwieldy at times. While managing
  biosecurity responses through traditional response structures utilizing State and local control centres
  may be entirely appropriate for some of the highly contagious diseases of terrestrial animals (think
  FMD), for aquatic animal diseases the need to have decision makers so remote from the affected
  area/farms may not be so critical, given the different pathways of disease spread (usually water
  related).
- Perhaps in part because of the various different layers of the response structure, communication with farmers was lacking in some instances. This was evident when BKD first arrived at 2IP, 3IP and 4IP and met with farmers who had not received any information from authorities and hence felt they were operating in an information vacuum. Prompt advice to all farmers in the area about basic biosecurity precautions (e.g. the potential risks of disease introduction from intake water) at the very earliest stages may have reduced their risks of infection.
- It is important to recognize that industry peak bodies (in this case APFA) may not have sufficient resources to manage the quantum of communication and meetings this capacity needs to be recognized and addressed earlier in the response process.
- Interpersonal interactions at the farm were also identified as problematic. Several farmers complained about BQ staff turnover they never knew when their site controller would change. Just when they may have "inducted" one person onto the farm and gained some proficiency in working with them, that person would be replaced by another and the whole process would have to be repeated. Even acknowledging the unprecedented scale of the response (for an aquatic event), a more stable roster process may have been able to reduce/prevent these problems.
- A large amount of stress for farmers arose due to the fact that many instructions to farmers were verbal and not backed up by written documentation. Indeed, several farmers did not receive any written documentation regarding testing results or even documents outlining why their whole farm was being chlorinated until half or most of it was already wiped out. The only thing worse than no information is misinformation, which often happened when verbal instructions dominated and the response strategy appeared to vary from day to day or hour by hour. Clearly this is not satisfactory and in the future it is important that relevant documentation is provided to farmers as promptly as possible and written (hard copy) situation updates are also provided to farmers on a regular, predictable basis. The urgency for eradication should in no way be used as an excuse to keep farmers in the dark.
- A fundamental of disease surveillance is that the chain of custody of samples should be complete to
  ensure that samples are collected from sites that are properly identified. Sampling should also be
  biased towards collecting diseased or suspect animals first, followed by random samples if no
  suspect animals are available. Sampling of prawns from feed trays biases samples towards healthy

feeding prawns, which is at odds to the objective of disease surveillance. Cast net sampling (ideally with a single cast net per pond to minimise risk of cross contamination), appears a good compromise in this regard, but all pond edges should always be observed first for animals exhibiting unusual behaviour or other signs of disease, and if present those animals should be sampled first before random sampling commences.

- Eradication strategies for disease incursions in prawn farms should always be flexible enough to allow emergency harvest to cooking on the infected property for any disease agent that is inactivated by heat (such as WSSV). During eradication whether the disease agent is inactivated by physical (heat) or chemical means (e.g. chlorine) should be irrelevant, it is the inactivation that matters, and indeed any method of inactivation which allows removal of biomass from ponds as part of the process (e.g. emergency harvest to cooking on site) will make the treatment of the pond water more effective and subsequent management of the pond contents far easier (e.g. carcass disposal occurs through normal processes).
- Control of birds or other mobile predators is absolutely necessary to reduce the chances of rapid movement of exotic diseases from infected premises to uninfected areas. There were several instances where scare ammunition was used when birds were actually in WSSV positive ponds, leading to spread of the virus rather than containment. It was clear at the coalface that live ammunition should always be available and bird control officers need to be prepared to use it when necessary to prevent disease spread whenever birds physically enter infected ponds.
- During a response to an exotic disease, access for BQ staff to all farms in the area at all times should not be negotiable. It is possible that failure to place bird control officers on 8IP during harvesting there and the inability of BQ staff to oversee the emergency harvest at 8IP ultimately lead to infection of 7ARP when hundreds of birds gained access to drain harvested WSSV positive ponds at 8IP, which were only 600-750 meters away from the then last remaining WSSV free farm on the Logan River.

### 4.2 Recommendations relating to the disease

- While the majority of research efforts on the Logan River have understandably been focused on surveillance to inform eradication efforts, during attempts to understand how the disease has been introduced and spread within and between infected farms, there have been opportunities to learn more about the disease itself.
- As much of the pond-level epidemiological information on WSD has been gathered in countries such as Thailand, Vietnam, Indonesia and India where the disease was introduced many years ago and the virus has become well established in wild crustacean populations (Corsin et al. 2001, 2003, 2005, Cullinan et al. 2013), the outbreak on the Logan River has provided a rare, perhaps even unique opportunity to examine the epidemiology of the disease in a new environment with naïve hosts under conditions of low environmental levels of virus.
- Under these conditions the spread of the disease in and between prawn farms on the Logan River appeared at first glance to be completely at odds with the circumstances described in regions where

WSSV has been long established in wild populations. In such cases, proximity to the river or sea is usually an important pond level risk factor, meaning that prawn ponds closest to river or sea tend to have the highest risk of infection (Corsin et al. 2001, 2005, Cullinan et al. 2013). The reasons why this is so are not entirely clear, but it is thought to be due to increased exposure to the virus via vectors such as crabs, birds or insects or though less obvious pathways such as movement of water containing free or particle associated virus through porous bund walls. But in most cases where WSSV has become endemic, it seems that massive environmental loads of WSSV can drown out potentially promising, yet subtle epidemiological risk factors such as transient WSSV positive periods in pond plankton, leading to indeterminate results (e.g. Callinan et al 2013).

- In contrast, in all but one of the farms on the Logan River, the index ponds were located furthest away from the river at the ends of intake canals. While a complete array of epidemiological data are not available, perusal of available records (Tables 1, 3) suggested that early pond stocking date was a risk factor in some farms, but not others, nor was pond water quality a consistent factor, nor food source or PL source. Instead, the major risk factor for farm infection appeared to be water intake from the river or Moreton Bay within the previous 7-15 days. Indeed, water quality data for the index pond at 1IP suggested a water intake occurred on 14<sup>th</sup> November (60 days post stocking), with clinical disease occurring 8 days later after the prawns experienced a transient reduction in water temperature at 62 days post stocking (Table 3). Reductions in water temperature are a well known risk factor for initiation of disease in populations of prawns already infected with WSSV (Corsin et al. 2005), suggesting that the virus may have been introduced into pond 12 at 1IP in a water intake around 14<sup>th</sup> November 2016.
- Besides recent water intake, the main risk factor for individual index ponds at all (except the last) infected farms appeared to be pond location, in particular those with takeoff of water from the downwind end of intake canals. Observations at night at 5IP suggested that this may be explained by non-random distribution of crustaceans or other potential vectors (phytoplankton, zooplankton, insect larvae) which occur in the intakes. At 5IP, the distribution of these was strongly influenced by wind direction (i.e. crustaceans and other potential vectors concentrated at the downwind ends of the intake canals), with predominately north westerly winds in the days leading up to clinical disease meaning the south easterly ponds on the main intake channel tended to be infected first. Corsin et al. (2001) observed that WSSV "must have entered infected ponds with water or some unidentified carrier in the water". The evidence on the Logan River tends to agree with this observation, suggesting that under conditions of such low environmental levels of virus (c. 0.1% prevalence in decapods), viral loads may need to be concentrated by wind forcing of particle associated virus or (presumably very small) vectors into certain downwind areas of intake canals prior to reaching the minimum viral threshold required to initiate infection of decapods once that water was taken into production ponds. Such phenomena would not necessarily be observed in regions where WSSV has been established for long periods, as such subtle processes would probably be drowned out by noise from a myriad of other pathways and variables.
- The only exception to the index pond pattern was in the last farm to be infected (7IP), which had not taken in water for over 2 months, remaining WSSV negative until a week after large numbers of birds were observed wading in WSSV positive ponds at 8IP, around 600-750 meters to the east

- (Figures 6, 7). In this case, it appears temporally and spatially plausible that the introduction of WSSV into pond 4 at 7IP was via movement of birds between 8IP and 7IP, especially as the birds from ponds 16 and 17 would have been disturbed when those ponds were chlorinated. In the absence of more detailed epidemiological information or eyewitness accounts, we may never know for sure about 7IP. However, it does appear statistically implausible that bird movements could explain the non-random transfer of disease between earlier farms from 1IP through 3IP, 4IP, 2IP and 5IP in the manner observed.
- Given that *P. monodon* appear so exquisitely sensitive to infection by WSSV (based on high infection prevalences in ponds and 100% prevalence in presumed escapee *P. monodon* in the wild, vs prevalences of c. 0.1% in all other species of wild crustaceans examined to date), they appear to be particularly well suited for use as sentinels either in production ponds (at normal commercial stocking rates with suitable risk mitigation including pond lining, crab fencing and bird netting), or held in sentinel cages on the Logan River itself. There is no doubt that there remains much to be learned about WSSV infections in the wild on the Logan River, and as testing continues and expands to plankton and other potential vectors we may yet be allowed an unprecedented insight into the epidemiology of the virus in the wild during this incursion.

### 4.3 Summary relating to the incursion

- The fundamental question that underpins this entire incident is how did WSSV get into 1IP in the first place? Biosecurity QLD has been considering four incursion pathways, namely:
  - 1. WSSV enters the Logan River via infected imported prawns being used as bait/burley
  - 2. WSSV is endemic in broodstock vertical transmission through PLs
  - 3. WSSV has been present in QLD waters for some time, or
  - 4. WSSV entered in imported feed/products.
- Of these, pathway 3 appears least likely, as if this were true, WSSV outbreaks would have been observed on the Logan River and elsewhere before November 2016. Pathway 2 also appears extremely unlikely as many farms outside the Logan River that were stocked by Rocky Point hatchery remain WSSV negative, and several of the farms affected on the Logan River obtained PLs from different sources. Pathway 4 appears equally unlikely, as sources of feed varied between farms and Ridley feed that was fed at 1IP immediately before the outbreak is extruded at temperatures of 85+°C for >30 min which would inactivate the virus (Ridley 2016), and those products are fed to many farms outside the zone that remain uninfected. Jim Thompson from BQ stated in a meeting on 2 February 2017 at Yatala that "The most likely pathway of introduction appears via imported prawns used as bait". I agree this is the most logical and likely pathway, for the following reasons.
- Introduction of WSSV via bait or burley would explain the detection of the single mud crab in the outlet canal of the then 7ARP on 23 December 2016, at a time well before nearby farms became

infected. The location where the crab was detected was near the road in an area frequently fished by recreational fishers.

- The bait and burley pathway has been consistently identified as a high risk pathway for dissemination of aquatic animal diseases by several risk analysts (Hine and MacDiarmid 1997, Durand et al. 2001, Hasson et al. 2006, Diggles 2011, Oidtmann and Stentiford 2011, Jones 2012). Furthermore, there is evidence that the number of people using prawns sold for human consumption as bait in Australia has increased (Kewagama Research 2007), representing increasing risk of WSSV introduction over time via this pathway (Oidtmann and Stentiford 2011). While the majority of green imported prawns sold for human consumption are probably used for their intended purpose, it is well known that some recreational fishers in Australia use imported prawns purchased from supermarkets as bait, because they are cheaper and perceived to be better quality (QLD Government 2006, Fishraider.com.au 2013, Fishing Victoria 2016).
- Despite biosecurity protocols requiring testing of 100% of shipments of frozen green prawns
  imported into Australia, there is evidence of a biosecurity failure allowing WSSV-infected frozen
  green prawns to transit through border quarantine in Australia on a regular basis, at least in part due
  to attempts by some importers to evade detection at the border by mislabelling high risk
  commodities and substituting known WSSV-free prawns for testing (Atkin 2017).
- The latest data from enhanced surveillance testing at the international border has resulted in a rejection rate of 70.3% (26/37) of green prawn shipments that are WSSV positive. This is slightly higher than the 66% batch prevalence detected by researchers in the EU who surveyed imported green *P. vannamei* prawns at British supermarkets (Bateman et al. 2012), but probably represents the "normal prevalence" of WSSV in commodity prawns today. The 70.3% rejection rate at the border under proper surveillance needs to be compared to the 16.3% rejection rate (73/448 consignments) reported by DAWR for shipments between May and December 2016. The difference in these two figures (70.3 16.3) is 54%, which is very close to the 50% prevalence at retail outlets alluded to by DAWR staff in phone conversations (B.K. Diggles, unpublished data). Thus, 50-54% represents the likely prevalence of WSSV infected prawns that cleared quarantine and were available to consumers at retail counters in Australia in the leadup to Christmas/New Year 2016.
- Since the Logan River crustacean fishing area closure has been implemented, fisheries officers have reportedly detected at least 6 groups of recreational fishers near the Alberton Boat ramp using imported raw prawns as bait. Of the 6 bait samples confiscated and tested, 2 (33%) returned "strong positive" results for WSSV infection. The fact that recreational fishers operating near the affected prawn farms were caught using WSSV infected imported prawns for bait was officially confirmed by the Federal DAWR in a media statement by Deputy Secretary Lyn O'Connell. So we can be confident that WSSV was being introduced into the Logan River by recreational fishers using imported prawns as bait or burley. While the less likely risk of bioterrorism (Jones 2012) in the

form of deliberate introduction of imported prawns into the intake of 1IP cannot be ruled out, it is probable that even if that occurred, there would be insufficient evidence available to make a successful prosecution.

- Viable WSSV has been recovered from crustacean tissues (including commodity prawns) frozen at both -20 and -70°C after months to several years storage and used to successfully infect susceptible crustaceans (Wang et al. 1998, McColl et al. 2004, Hasson et al. 2006, Biosecurity Australia 2009, Bateman et al. 2012, RM Overstreet, personal communication, Nov 2009). Viral loads of between 10<sup>8</sup>-10<sup>10</sup> viral copy units/g tissue occur in diseased emergency harvested prawns (Oidtmann and Stentiford 2011). Removal of the head section does little to reduce WSSV viral load on a per weight basis, as viral load in individual prawns is nearly identical in either heads (49% of total virus) or tails (51% of total virus) (Durand et al. 2003), and viable WSSV can still be detected in commodity tails (McColl et al. 2004, Hasson et al. 2006, Biosecurity Australia 2009, Bateman et al. 2012), with the virus load of the peeled shell representing approximately 55% of the total viral load remaining in the tail (Durand et al. 2003). Hence full processing of green prawn products only reduces viral load by around half, which is not at all sufficient to prevent establishment of infections in susceptible species (Bateman et al. 2012).
- Recreational fishers were subsequently observed fishing with WSSV positive imported prawns in the Logan River near the intake canal of 1IP, and there is evidence of regular fishing in that intake canal itself, including right up to within a few meters of the intake pipes. These canals have limited water exchange, low dilution factors, and are frequented by large numbers of potential hosts and vectors (crabs, prawns, plankton) and hence they represent semi-isolated environments that are perfect for establishment of WSSV infection in wild reservoir hosts and vectors. All that is needed is for northerly winds to concentrate infected vectors at the end of the canal then have that water pumped from the intake up into the intake channels at 1IP where wind forcing would direct and concentrate particle associated virus or vectors near the end of the channel near index pond 12 and pond 13. Around the time of the suspected introduction (Nov 14) water intakes into pond 12, but not pond 13 (Luke Rossmann, personal communication to BKD, 6 Jan 2017) could explain disease emergence in that pond first.
- Once the disease outbreak began in pond 12, large amounts of virus could have been released into the settlement pond and 1IP outlet canal during pond flushing activities between 25 and 30<sup>th</sup> Nov 2016. Local rainfall around this time may have further increased risk of virus being introduced into the outlet canal and infecting wild crustaceans there (Figure 4). The 1IP outlet canal remained open to the Logan River until around 4<sup>th</sup> December, potentially allowing a week or more for virus and/or infected hosts to escape into the Logan River, which would explain the infection of 3IP (which was still taking in water on the 4<sup>th</sup> December) and detection of WSSV positive *Metapenaeus* spp. and *Acetes* spp. taken from the river near the outlet to 1IP on 5<sup>th</sup> December in trawl sites #2 and #3. As 2IP ceased water intake on the 2<sup>nd</sup> December, it is possible that this farm was exposed to lower levels of virus which allowed it to remain free from disease until 12<sup>th</sup> December. Once in the river,

other farms downriver would likely be exposed to WSSV via free virus, particle associated virus or vectors in their intake water, as previously described (Figure 5).

- The strong possibility that this disease incursion was caused by use of imported prawns as bait signals an urgent need to revise the prawn IRA and reassess this and other potential pathways of aquatic animal disease introduction into Australia. The IRA has now not only failed, it is simply out of date. The risk profiles for diversion of prawns and other imported seafood products to bait and burley have either changed or were not properly identified in the first place, and new phytosanitary information is now available on risks related to WSSV and many other emerging (post-2009) diseases (Table 5) in imported prawn commodities (see papers by Overstreet et al. 2009, Ma et al. 2009, Stentiford et al. 2009, Oidtmann and Stentiford 2011, Reddy et al. 2011a, 2011b, Bateman et al. 2012, Stentiford et al. 2012, Stentiford 2012, Jones 2012, Shields 2012, Behringer 2012, Lightner et al. 2012, Tran et al. 2013a, 2013b, Reddy et al. 2013, Nunan et al. 2014, De La Pena et al. 2015, Cowley et al. 2015, Thitamadee et al. 2016, Bateman and Stentiford 2017, amongst many others).
- The reason why Australia has not yet got some of these new diseases is pure luck. For example, the toxin related components of the bacterium that causes Acute Hepatopancreatic Necrosis Disease (AHPND) appears to be inactivated by freezing, which is fortunate otherwise Australia would likely be included in the estimated \$5 billion US annual global losses experienced by overseas prawn producers due to AHPND (see Tran et al. 2013a, 2013b, Chamberlain 2013, Thitamadee et al. 2016, Li et al. 2016).
- The fact that some States (Western Australia and South Australia), quickly moved to protect their environment and valuable fisheries and aquaculture industries by implementing controls on movements of uncooked prawn products from the Logan River area to try to prevent WSSV incursions into their own waters, highlights a remarkable inconsistency in what is considered an Appropriate Level of Protection (ALOP) by State Governments in Australia, compared to the Federal Governments current (pre-interim closure) position on imported prawn products. Having stricter controls requiring cooking of Australian prawns moved domestically from WSSV positive regions, yet still allowing uncooked imported prawns entry at the border from WSSV positive regions overseas is an extraordinary situation that highlights exactly where the real risks lie.
- Given the scale of the biosecurity breaches revealed at the border, and the potentially severe consequences of introduction of exotic diseases to Australia's fisheries and aquaculture industries, relying on luck is simply not good enough. Only a comprehensive review and full update of the prawn IRA (and the resulting biosecurity protocols implemented at the international border) is acceptable, so that Australia's environment, seafood industries, and food security for future generations are given the full consideration and attention the people of Australia deserve and demand.

Table 5. List of some of the diseases of prawns that were not included in, or have emerged since the 2009 Import Risk Assessment (data collated only from Thitamadee et al. 2016, Li et al. 2016, Bateman and Stentiford 2017 and is not an exhaustive list).

Disease name	Date emerged	Disease agent	Mitigated by existing phytosanitary measures?
AHPND	2009 (China)	Bacterium w. toxic plasmid	Yes
Secret Death Disease	?	Possibly AHPND or mixed aetiology	?
Empty Stomach Disease	?	?	?
Aggregated transformed microvilli (ATM)	2009 (China)	Vermiform gregarine-like bodies	?
Covert Mortality Disease (CMD)	2009 (China)	Nodavirus	?
Hepatopancreatic microsporidiosis	2009 (Thailand)	Microsporidian (Enterocytozoon hepatopenaei)	?
Hepatopancreatic haplosporidosis	2009 (Indonesia)	Unnamed haplosporidian	?
New strains of YHD	2013 (China)	Okavirus	?
Pandalus montagui bacilliform virus	2007 (North Sea)	Nudivirus	?

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# Appendix 1. WSD outbreak situation report #1. 2 Dec 2016

I was on the ground at the affected farm between 1 pm and 7.30 pm yesterday (2<sup>nd</sup> Dec) and worked together with farm staff and Biosecurity QLD officers to inspect water flow at the nearest unaffected farm before entering the infected area to collect information and document the situation on the ground with the objective of gathering data which will allow updating of relevant Aquavet manuals sometime in the future, and to assist farm staff and industry with biosecurity advice upon request. Below is a map of the farm layout and its relationship with nearby farms.

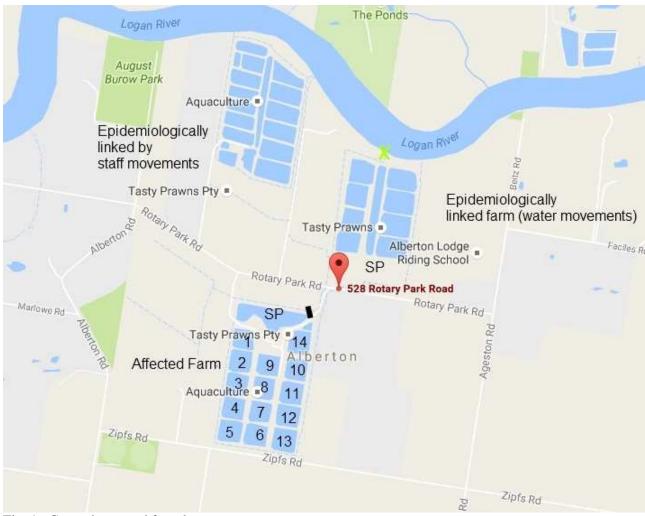


Fig. 1. General area and farm layout.

Some of my notes and observations from the day included the following:

- 1. Farm staff inform me that the outbreak was initially observed in Pond 12 (Fig 1), then has subsequently spread (based on my pondside observations) to ponds 11, 13, 10, 8, 7, 5 and 4. I am uncertain re: the status of ponds 9 and 6, but the prawns in ponds 3, 2 and 1 still appeared healthy as of 6.30 pm yesterday.
- 2. Biosecurity QLD have started treating affected ponds, starting with pond 12 (index case), then pond 11 which were treated with chlorine (>30 mg/L) yesterday evening. A total of 13,000 L of liquid chlorine (13% active shipped in a truck) was required to achieve 30-40mg/L in pond 12, while 17 x 1000L drums of 10% active chlorine (hypo 10) was being introduced into pond 11 as I left.

Biosecurity QLD were monitoring the decay rate of active concentrations of chlorine overnight (they are targeting >5 mg/L active at 24 hours post application), before treating the next pond (probably 10 or 13). Availability of sufficient chlorine (currently approx 40,000 L on site) over the weekend is restricting their ability to treat more ponds in the short term, but another 100,000 L should be available on site sometime Monday.

3. Biosecurity QLD have established a control area at the western entrance to the infected premise and have used bulldozers to form an earthern bank to block any effluent from leaving the settlement pond on that property (SP in Fig 1.). However upon speaking with staff from the farm, there is a possibility that decapods in the effluent drain that flows from the infected premise under Rotary Park Rd and past the second farm into the Logan River (Fig 2) were exposed to some water from the affected ponds as initial advice to farm staff (late last week) was that an unrelated disease may have been involved and that pond flushing might help. Obviously this situation has changed with the diagnosis of WSD and there is no longer any discharge through the earthen bank.

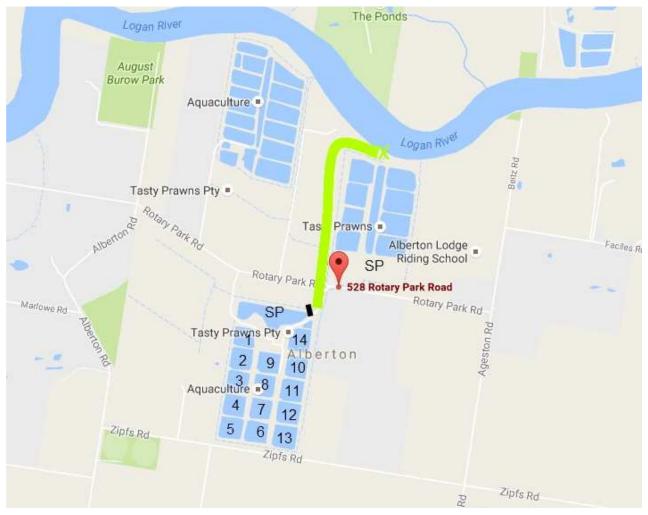


Figure 2. Location of effluent drain (highlighted) from earthern bank (black line) on infected farm draining under Rotary Park Rd to settlement ponds of farm to north then to Logan River. Effluent drain can be blocked at X.

4. There may have also been flow in these effluent drains after recent rains in the past week (as they also drain nearby cane farms). As of last night (7 pm) the pipe connecting the northern end of the effluent drain to the Logan River was not blocked off where it enters the Logan (see X in Fig 2). This effluent drain directly connects the infected farm with the settlement ponds of the farm nearest

to the north, making them epidemiologically linked via water flow. To me it would appear prudent to block this off and treat the effluent canal (highlighted in Fig 2) as a matter of some priority to minimise risk of potential release of WSSV exposed decapods into the Logan River. Farm staff and weather reports should be consulted before doing this though as with the effluent drain blocked off, heavy rains could cause backflow from cane farms. With limited chlorine availability until Monday, it will be up to Biosecurity QLD to determine how to allocate the available chlorine/human resources to do this.

5. Farm staff initially remarked to me that they thought the disease may have been a slightly different type of WSD, as the prawns looked different to textbook cases. I can confirm, however, that the prawns were probably simply dying too fast in the initial stages to form the large coalescing spots we see in textbooks, as a small proportion of those that were surviving longer in pond 11 displayed classical gross signs (Fig 3). For future industry educational purposes, some photos and videos of clinically affected prawns at the pond edges were taken and some fixed samples collected (davidsons fixative, samples remaining on site for now until cleared by Biosecurity QLD).



Figure 3. Gross signs of WSD in a moribund *P. monodon* from pond 11.

6. Farm staff appear to be holding up well, but are understandably tired and under resourced. Biosecurity QLD staff appear well trained (although mostly terrestrial biosecurity experience on site) and there appears to be sufficient people on site to control birds, administer chlorine etc, but there are large numbers of insects (particularly grasshoppers) in the pond surrounds and the algae on the pond edges where large numbers of dying prawns end up – these insects could also act as

mechanical vectors as virus levels in the water are likely to be very high once ponds start to go under. Spraying for insect control should be an option worth considering as the surrounding area has lots of water in drains etc and this could be one avenue of spread that is being overlooked.

**How can industry assist in a practical way?** In my opinion, there needs to be better control of vehicle movements and human activities in what should be the control area on Rotary Park road. After leaving the farm linked by effluent water, I witnessed some people fishing with castnets for prawns (*Metapenaeus* and banana prawns are their alleged normal captures) to use as live bait elsewhere (Fig 4). I inspected their operation and confirmed they had caught no prawns over a period of 10 min then informed Biosecurity QLD officers who ensured the people moved on.



Figure 4. A members of the public castnetting the intake canal for wild prawns for use as bait on Friday 2 Dec. For reference the white pajero is parked exactly at the east entrance to the infected premise (where the red placemark is in Fig 1).

Until such time as testing proves otherwise, it should be assumed that the intake canals in the control zone could potentially harbour infected hosts, and clearly having members of the public fishing in the control zone is a risk (where is the control?). While it is important to exclude unnecessary people from the infected area, it may free up some Biosecurity QLD staff if a responsible industry person could park on the road in the control zone near where the red placemark is in Fig 1 and keep an eye out for people who stop there to fish the intake drain. That person could inform Biosecurity QLD staff at the controlled entry point by phone whenever people turn up to fish so the fishers could be asked to move on.

# Appendix 2. WSD outbreak situation report #2. 5 Dec 2016

I was back in the affected area today to survey the intake of affected farm 1, and inspect Ian Rossmans epidemiologically linked farm 2 directly to the north, but was asked by Ian to instead first visit D&S Farm (Farm 3, run by Dan and Simon). Upon pulling up at D&S Farm at around 9.30 am I was met at the gate by Dan who informed me he had just found some sick prawns. I was taken to pond 2 (see Fig 1) and observed around half a dozen moribund prawns at the pond edge. Dan removed one and it showed the same clinical signs as some of the prawns I had observed from the affected farm on Friday (lighter cuticle discolouration etc.). Upon seeing this I abandoned the planned visit to farm 2 for the day and went to the control point at Farm 1 (approx 500 meters to the south) and alerted Peter Mowett and rang Steve Wesche to inform them of the situation at farm 3 by 10.15 am. I then returned to farm 2 to conduct a site inspection to learn about the pond layout, as well as intake and effluent flows and to provide support and advice to staff about biosecurity related issues.

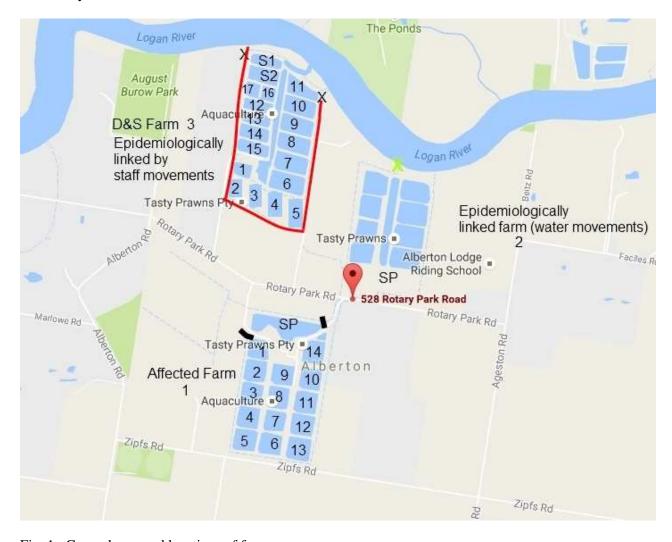


Fig. 1. General area and locations of farms.

I plan to continue the inspection of farm 2 tomorrow afternoon, but in the meantime some of my notes and observations from today included the following:

#### FARM 3

- 1. Staff at Farm 3 informed me that they felt they have been operating in somewhat of an information vacuum, they were unsure what was happening at farm 1 and felt they had received no firm advice from authorities on what to do. They also informed me that they had been taking in water on a regular basis, the last time they did this was less than 24 hours earlier on Sunday night (4<sup>th</sup> December). I informed them that appeared to be a high risk strategy considering the current situation nearby, however they said due to leaking boards on several of their pond outlets they have to pump regularly or they lose too much water, and no one had told them that pumping water was a high risk activity under the circumstances.
- 2. They also informed me that the samples taken for testing last Thursday (including from pond 2) were taken from feeding trays, so in effect were non-random and biased towards reduced sampling sensitivity because only feeding prawns would have been tested. Assuming these tested as negative, and that pond 2 will subsequently test positive for WSSV (see figures 3,4 below), given that one of the first signs of WSD is prawns going off their feed, it would appear prudent that in the future a more targeted sampling approach is necessary random cast netting would appear to be a better option to improve sampling sensitivity (provided cast nets are properly sanitized between ponds or are bulk purchased and allocated to one or two ponds only on each farm).
- 3. Upon inspecting the intakes and drains of farm 3 I noted that staff, suspecting WSD in pond 2, had blocked off both exits (X) of the main drain (red coloured line in Figure 1) by 11 am to prevent water flow to the Logan River which borders the northern part of their farm. Water is usually pumped from short intake canal into a main supply channel along the middle of the farm using 2 screened pumps. I investigated 50 or so meters up the intake canal and witnessed yabby holes and apparently healthy toadfish and fiddler crabs, but no dead crustaceans. However, the staff on site did report seeing 2 dead crabs (species unknown) in their main supply channel recently. I inspected the supply channel and did not see any dead crustaceans, but did witness 2 ducks on the water.
- 4. I inspected the circumference of every pond except number 2 and did not observe any moribund prawns or abnormal prawn behaviour in any other pond. I did, however, observe a reasonable number of grasshoppers (Figure 2) in the pond surrounds and at the waters edge. Upon finally visiting pond 2 again (at 2.45 pm) it was evident that more moribund prawns were appearing along the pond edges, particularly the upwind and downwind edges. By this time several prawns were swimming on the surface and exhibited reddened carapaces and other gross signs consistent with the WSD that I had witnessed at farm 1 on Friday (Figure 3). Farm staff had sent some samples of moribund prawns away for testing earlier in the day and had completed sampling of all ponds again to better ascertain prawn sizes and disease status of each farm pond in case an emergency harvest was possible. Before sending the samples off I was shown one prawn from pond 2 which appeared to have classical textbook coalescing white spot lesions (Figure 4).
- 5. In the absence of any basic biosecurity measures on site I advised against any movements of staff and equipment near pond 2 and helped staff arrange foot and hand washing baths at the farm entrance and other critical areas using a drum of benzalkonium chloride that was on site, at around 5000 mg/L for footbaths and 1000 mg/L for washdown of solid surfaces. A dose of 200 mg/L for 30 min is reported to be virucidal for WSSV so higher doses would be required due to reduced contact time for hand and foot washes. It was noted that farm staff were trying to undertake bird control where they could, but there is no guarantee they will have the resources to do this long term. Indeed the behaviour of farm staff was exemplary in the face of an extremely stressful situation, and they deserve as much support as they can be given ASAP.
- 6. I consider the situation at farm 1 to be **extremely urgent**. There is a more than reasonable suspicion that WSSV infected prawns are present in pond 2, and the infected pond is much much

closer to the river and has much less capacity to control pond effluent. Indeed, once the effluent channel was blocked at 11 am today it began to fill at a noticeable rate (Figure 5) suggesting that treatment of both Pond 2, possibly adjacent ponds (1 and 3) and the effluent channel to the west of the property are urgently required ASAP whenever the next batch of chlorine is available, so that this possibly dire situation can be better managed.

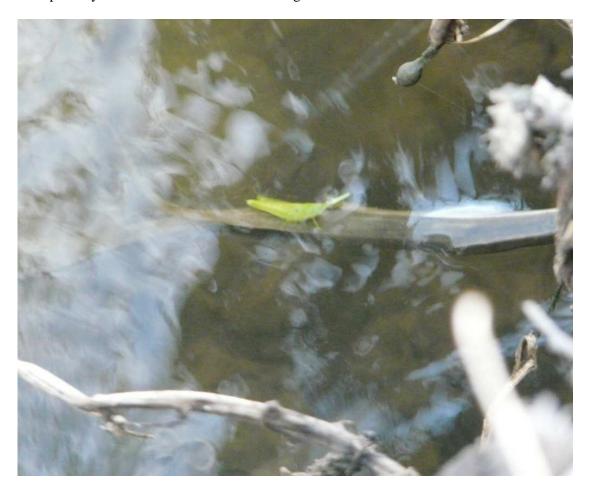


Figure 2. Many grasshoppers were evident in the water and at the waters edge at farm 3, as they were at farm 1.



Figure 3. Moribund prawn swimming at the water surface at pond 2 in farm 3 at 3 pm— note reddish discoloration of the carapace.



Figure 4. White spots on carapace of a prawn sampled from pond 2 by farm 3 staff. This prawn was included in the chilled sample sent off for testing on the Monday afternoon (c. 4.30pm).



Figure 5. Northern end of western effluent channel at farm 3, looking south. The canal was blocked here at the northern end around 11 am on Monday 5<sup>th</sup> and is beginning to fill up at quite a rate. This will need to be decontaminated before being allowed to empty again into the river.

### FARM 1

- 7. At the end of the day (6.15 pm) I left farm 3 and attended the control point at farm 1 to update myself on happenings there since Friday. It was clear they had been focusing on the effluent drain (which had been blocked and treated with hydrogen peroxide over the weekend) and today the sedimentation pond (SP in Figure 1). A new earthern wall had been made upstream of the sedimentation pond, presumably to prevent dilution of the chlorine in the pond itself. I was informed by Biosecurity QLD staff at the control point that the settlement pond required nearly all of the available chlorine on site (a large proportion of the approx 100,000 L available) in order to achieve the targeted dose, which meant that there was none left to send to farm 3 late in the day. This is unfortunate, as described above the newly suspected WSSV infected pond at farm 3 is much closer to the river and has much less capacity to control pond effluent all the good work at farm 1 could be to no avail if the situation at farm 3 is not bought under control ASAP.
- 8. Biosecurity QLD staff at the farm 1 checkpoint also confirmed that ponds 12, 11, and 13 have been treated with chlorine, representing one additional pond treated since Friday evening. However, prioritizing treatment of the effluent drain and settlement pond is logical as it should provide a reasonable buffer against virus making it into the river (all the more reason to prioritise treatment of Farm 3 ASAP today (morning of 6<sup>th</sup> Dec), see above). Data from the ponds treated to date was reported to be encouraging, with peak concentrations obtained around 50 mg/L in each pond with between 15 and 25 mg/L generally remaining after 24 hours, which should be an effective virucidal dose.

- 9. I then entered the infection zone at farm 1 at 6.30 pm to do an inspection and noted no obvious mortalities in Pond 14, but that mortality rates had increased markedly since Friday in ponds 10, 9, 8 and particularly 7, with moribund prawns also observed in ponds 5 and 4. I am uncertain re: the status of pond 6 as it is murky, and the light was fading when I viewed ponds 3, 2 and 1 but nevertheless I could not see any moribund or dead prawns in the latter 3 ponds, as was the case on Friday. So it appears that the disease is progressing rapidly in ponds 7-10 which are all adjacent to the index pond (12), so it looks like horizontal movement through the farm from east to west over time. Upon leaving in the dark (7.30 pm) I noticed several canetoads hopping around the ponds which could be another potential mechanical vector in the absence of fencing between ponds.
- 10. I also noted there still needs to be better control of vehicle movements and human activities in what should be the control area on Rotary Park road. I went down to where the road intersects the intake of farm 1 (near where the red placemark is in Fig 1) at 6.10 pm (dusk, prime fishing time) and saw no one present to stop people fishing in the intake. As mentioned previously, until such time as testing proves otherwise, it should be assumed that the intake canals in the control zone could potentially harbour infected hosts, and clearly having members of the public able to fish in the control zone is an unnecessary risk (where is the control?).

# Appendix 3. WSD outbreak situation report #3. 8 Dec 2016

This is a summary of developments on Rotary Park Rd since my last report of 5<sup>th</sup> December 2016.

## 6<sup>th</sup> December 2016

On Tuesday 6<sup>th</sup> December around 9.45 am I was phoned by Dan who informed me that signs of WSD had been observed in prawns on farm 3 (IP3) not only in pond 2 (where clinically diseased and dead prawns were evident yesterday), but also in pond 3, which would have been exposed to aerosol effects from pond 2 under the prevailing north-NW winds. I was asked to attend and assess the situation. I arrived in the area at 12.45 pm but before entering any infected premises I firstly visited farm 2 to investigate its layout and view the health of the stock.



Fig. 1. General area and locations of farms, including pond numbers.

My notes and observations from farm 2 on 6<sup>th</sup> December included the following:

- 1. the prawns on farm 2 are visibly smaller than on the other two affected farms
- 2. several egrets were observed hunting in the settling pond and a pair of whistling kites was present soaring over the intake pipes.

3. Prawns in all of the ponds appeared healthy – no signs of abnormal behaviour or disease were observed.

After completing inspection of farm 2 at 2.15 pm and routine between farm decontamination (decontaminating footwear, camera, phone, hand washing and replacement of boots, outer clothing etc which I do before entering each site) I moved on to farm 3 at 3.30 pm to observe the health of prawns in the ponds there and be present when chlorination of WSD affected ponds started. My notes and observations from farm 3 (IP3) on 6<sup>th</sup> December included the following:

- 4. Upon entering IP3 I viewed pond 3 with the suspected WSD noticed earlier that morning. I observed abnormal behaviour including low numbers of lighter coloured moribund prawns swimming at the water surface, with occasional erratic flicking along the pond edges.
- 5. At pond 2 WSD had progressed rapidly since yesterday morning with significantly higher numbers of dead and moribund prawns (3-5 per meter) visible along the pond edges, mainly along the upwind pond edges with fewer on the downwind edge.
- 6. At approximately 3.45 pm a chlorine tanker truck arrived with enough chlorine to treat 1 pond only. After discussion between Dan, David Mann and myself, it was decided to decontaminate pond 3 in order to reduce chances of spread of the disease to ponds 4 and 5 immediately to the east. Decontamination of pond 3 began at approximately 4 pm.
- 7. After routine on-farm decontamination (footbath, handwash) a Biosecurity QLD officer (Laurie) accompanied me as I inspected the remainder of farm 3 to determine if the disease had spread to other areas of the farm. This inspection found no noticeable changes to the status of any other ponds on site compared to Monday 5<sup>th</sup>, except for noting that there were large numbers of grapsid crabs in pond 16 and a legal sized mudcrab was observed in pond 6. I also investigated 50 or so meters up the bank beside the intake canal and witnessed yabby holes and apparently healthy toadfish and fiddler crabs, but no dead crustaceans. Laurie noted that high quality polarized sunglasses were a necessity in order to properly observe pond inhabitants.
- 8. After observing nothing new in all other ponds we arrived back at pond 3 at 6.15 pm, noting that in the intervening 2 hours the chlorine had been effective in killing large numbers of prawns. Others showed signs of stress including jumping out of the water and onto the bank where many subsequently died without further exposure to chlorine, hence representing a potential risk of spread by birds of the later are not well controlled.
- 9. The site inspection was completed by 7 pm at which time I exited the facility for home where routine between-farm decontamination was repeated.

### 7<sup>th</sup> December

I did not attend the area on Wednesday 7<sup>th</sup> December but around 5.45 am I was phoned by Dan at farm 3 who informed me that chlorine had finally been introduced to pond 2 at 4 pm that day, but that the effluent drains surrounding the farm had still not been treated, and continued to fill. He also informed me that the prawns in pond 1 immediately adjacent to pond 2 till appeared normal.

## 8<sup>th</sup> December

On Thursday 8<sup>th</sup> December I was asked to attend the area and assess the situation on farm 3 (IP3) and also the situation with the intake to farm 1 (IP1). En-route I was asked to collect 15 cast nets to facilitate more intensive sampling (at some later date) of the apparently unaffected ponds at IP3 as well as the intake to

farm IP1. I arrived at IP3 at 2.00 pm with 16 cast nets (7 or 8 foot drop, 1 inch mesh) which are now on site for use as/when required.

My notes and observations from 8<sup>th</sup> December included the following:

- 10. I was informed by farm staff at IP3 that they arrived on farm early this morning and noted that no bird control staff were present, resulting in many birds being observed around ponds 2 and 3 that were decontaminated the previous 2 days.
- 11. A significant development occurred approximately 2.30 pm when Biosecurity QLD staff advised farm staff and myself that WSSV had been detected in wild crustaceans collected adjacent to the nearby Alberton boat ramp (BR in Figure 1). GPS co-ordinates show the boat ramp is around 560 meters from the farm gate at IP3 and probably only around 200 250 meters from the nearest settlement pond on the same property.
- 12. After this news I inspected the circumference of every pond on IP3 and did not observe any dead or moribund prawns or abnormal prawn behaviour in any pond (except for in the decontaminated ponds 2 and 3). A bird control staff member who I interviewed along the way (at pond 11) confirmed that no one was conducting bird control at IP3 overnight between around 10 pm and 6 am. The inspection was completed by 4.30 pm.
- 13. A total of 15 cast nets were then bought on site at 4.45 pm to ensure they were ready when needed, however when a farm is under a biosecurity order, movement of samples off site for testing requires Biosecurity QLD consent. Any samples taken for diagnostics would also need to be handled/fixed in an appropriate manner, especially if they are going to be stored on site for any length of time before being delivered to AAHL. Because of this, and as well as other issues such as the need for chain of custody for samples, I strongly suggest that any experimental or surveillance sampling from IP3 (and the intake of IP1 for that matter) that is requested by industry should be done in full collaboration with Biosecurity QLD and that the methodology of any experiments that may be conducted on IP3 should be agreed to by both parties before proceeding.
- 14. After completing tasks at IP3 and routine between farm decontamination (decontaminating footwear, camera, phone, hand washing and replacement of boots, outer clothing etc which I do before entering each site), I moved on to view the intake for IP1 (green line in Figure 1) at 5.15 pm.
- 15. I parked near the intake where it meets the road at 584 Rotary Park Rd and interviewed a local resident about the frequency of fishing activity in the intake. He informed me that it was not unusual to have 3 to 4 groups of people a week fishing in the intake, many with cast nets, and that around 2 weeks prior Dept. of Fisheries may have caught some people for illegal fishing in the intake. Another local resident then pulled up to inquire what I was doing and asked whether I had been fishing, showing that local surveillance of fishing activity on the road near the inlet has increased.
- 16. I could not ascertain whether the intake was closed or open to the river from my viewpoint on the road, so employed the help of Ian Rossman who took me to the intake entrance (via farm 2) in his car at 6.00 pm. I managed to ascertain that the intake was indeed closed near its entrance (X at northern end of green line in Figure 1, see also Figure 2) where both of the 2 flood gates had been closed by Ian since late pm on Friday 2<sup>nd</sup> December. Normally one of the hinged gates is open, allowing water to enter the intake canal, but not drain back out, meaning that the intake to IP1 represents a semi-enclosed sub-habitat of the Logan River. I assisted Ian in closing off the overflow pipe with reinforced plastic at around 6.15 pm (Figure 3) to prevent water from escaping if it rains. Since the intake has been closed since Friday 2<sup>nd</sup> December it is well worth intensively sampling it by castnetting or other random sampling means in order to improve epidemiological understanding of the origin of the index outbreak in IP1.



Figure 2. Location where intake of IP1 is blocked by dual floodgates (right arrow), while the overflow pipe is evident to the left (left arrow). The intake was blocked on  $2^{nd}$  December.



Figure 3. Reinforced plastic was placed over the overflow pipe (arrow) to prevent water escape.

17. After completing inspection of the intake I returned to the control point at IP3 to retrieve my other set of gumboots that I had left at the gate (which were missing, obviously borrowed!). So instead I observed that chlorination of the main effluent drain (red coloured line in Figure 1) began today shortly after a chlorine truck arrived at 7.10 pm. David Mann informed me that they had essentially finished dosing IP1 so were now positioned to focus more attention on IP3. With the assistance of IP3 staff, chlorine was observed being applied starting from the north west corner (blue line in Figure 1) from around 7.20 pm. I left the control point at IP3 at 7.30 pm and returned home where routine between-farm decontamination was repeated.

# Appendix 4. WSD outbreak situation report #4. 11 Dec 2016

This is a summary of developments along the Logan River since my last report of 8<sup>th</sup> December 2016. For clarity I have reverted to the Government nomenclature for identifying infected premises (e.g. 1 IP) instead of the codes I used in previous reports (e.g. IP 1).

## 9<sup>th</sup> December 2016

1. On Friday 9<sup>th</sup> December during the industry phone conference we were informed that all of the ponds in 1 IP had been chlorinated as of late PM on 8 December. We were also informed that 3 ponds at 4 IP had been diagnosed as PCR positive for WSSV, and that at 5.30 am that morning Pond 4 at 3 IP (Figure 1) had been chlorinated by unsupervised truck drivers and several other chlorine trucks were parked outside 3 IP ready to continue decontamination of non PCR positive, non-clinically diseased ponds at that site. It was suggested during the phone hookup that these trucks could be better utilised by directing them to known infected (PCR Positive) ponds at 4 IP. Subsequent conversations I had with farmers at 4 IP disclosed that the prawns in these infected ponds (1, 3 and possibly 2) began to show clinical signs of disease on the evening of 8 December 2016. We were also informed that movement controls had been implemented as of 8 December by Department of Agriculture and Fisheries in an attempt to prevent commercial and recreational fishers from moving known carriers of the virus in the area of the Logan River (Figure 2).

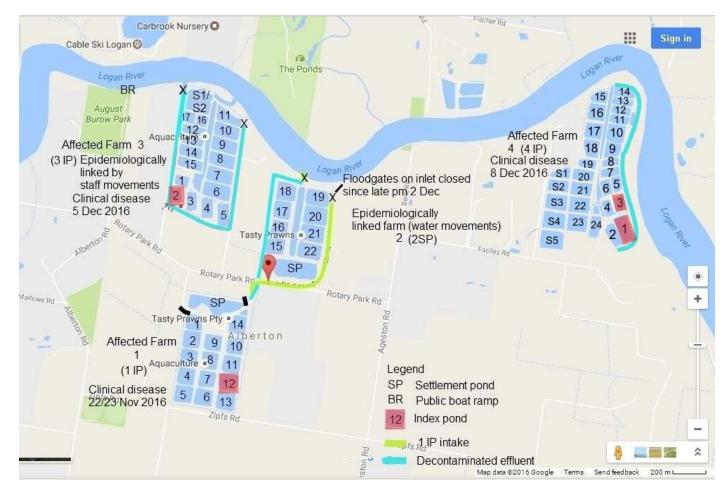


Figure 1. Locations of farms, including pond numbers and location of index ponds at each site.

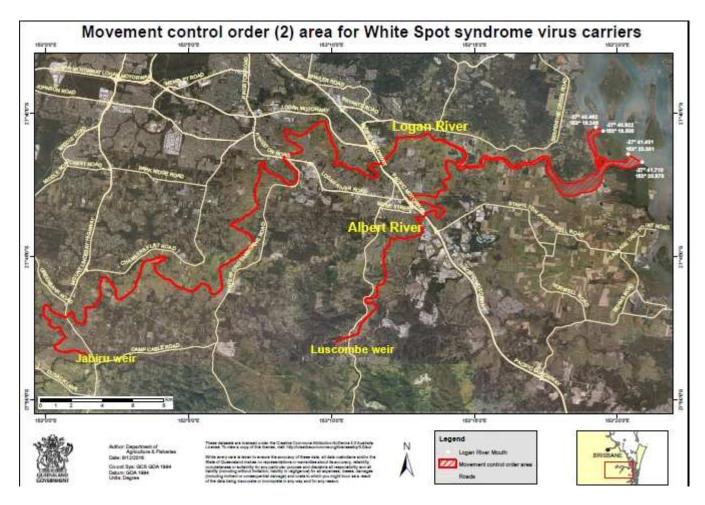


Figure 2. Map of movement control area declared by Dept. of Agriculture and Fisheries preventing fishing and restricting movements of all known WSSV carriers, including crustaceans (crabs, prawns, etc.) and polychaetes.

### 10<sup>th</sup> December 2016

- 2. On Saturday 10<sup>th</sup> December I was contacted by the owners of 4 IP who confirmed that 3 PCR positive ponds at 4 IP had been chlorinated vesterday (9 December) starting within 12-16 hours of the prawns in ponds 1 and 3 beginning to show clinical signs of disease. As for all of the other infected farms, the ponds on 4 IP that showed clinical signs of disease first and/or were identified by testing as being WSSV positive first were those ponds that were furtherest away from the intake pumps (Figure 1). This consistent picture throughout all 3 farms infected so far suggests the spread of the disease between farms is not random or caused by movements of birds or human activities, but instead is highly likely to be associated with pumping in virus (or more likely, hosts infected with the virus) from the river. A plausible hypothesis might be that ponds nearest the water intakes are receiving water that is in the intake/supply canals for a shorter time period, resulting in prawns being exposed to lower viral titres (virus/vectors would be diluted by higher water flows) and/or better water quality. In contrast, the ponds furtherest from the intakes may be taking water that has higher viral titres due to less water flow concentrating virus and/or vectors (e.g. jelly prawns Acetes spp.) at the very ends of the supply canals where water flow is least and water quality is likely to be reduced. This hypothesis may also be supported by the fact that Farm 2, which has not pumped water from the river since Friday 2 December, remains uninfected at this time despite being surrounded on all sides by infected farms. Other notes from 10th Dec included:
- 3. Limited experimental emergency harvesting commenced at 3 IP, but it was found that the prawns were relatively small and it may not be worth the effort.

4. That Ponds 1, 2, 3, 4, 5 and 6 from 4 IP had been chlorinated, together with the effluent discharge channel to the east of the property (Figure 1), resulting in the deaths of large numbers of fish (mainly mullet, eels, bream, and forktailed catfish).

## 11th December 2016

- 5. On Sunday 11<sup>th</sup> December I undertook a visit to the affected area in order to inspect the layout and status of 4 IP as well as collect some ethanol fixed samples (n = 60 randomly sampled prawns per pond using a castnet) from selected ponds at 3 IP so that samples could be archived (for later epidemiological studies) on site prior to the ponds being chlorinated.
- 6. Prior to visiting the farms, however, I first inspected Gem Bait and Tackle to observe whether they were still selling fresh locally caught chilled and frozen prawns despite the movement control area restrictions. I arrived at the tackleshop at around 11 am and almost immediately identified large numbers of locally caught wild crustaceans with clinical signs of WSD, including banana prawns (Figures 3, 4), greasyback prawns (*Metapenaeus* sp.) (Figure 5), river shrimp (*Macrobrachium* sp.) (Figure 5), and palaemonid shrimp (Figure 6), suggesting an epizootic in the river.

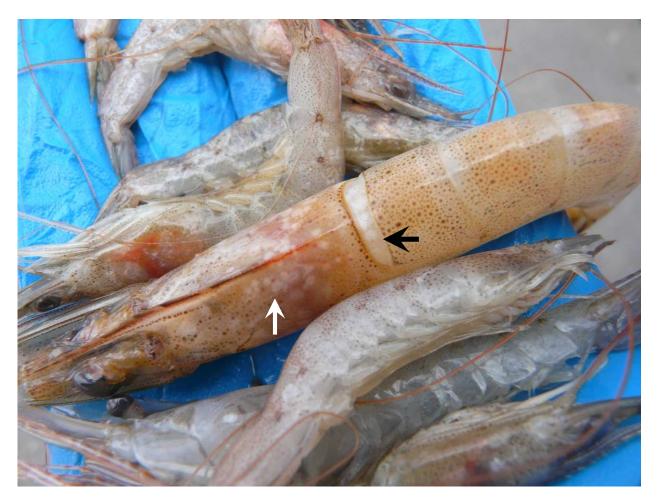


Figure 3. Mixed locally caught prawns being sold as bait on the morning of 11th December. Note signs of clinical WSD on the larger yellow banana prawn (white arrow), including lesions on the arthrodial membranes (black arrow). Note: testing of these samples gave indeterminate results (WSSV positive for some tests, but not others)



Figure 4. Signs of clinical WSD on a locally caught banana prawn being sold as bait on the morning of 11th December. Note: testing of these samples gave indeterminate results (WSSV positive for some tests, but not others)

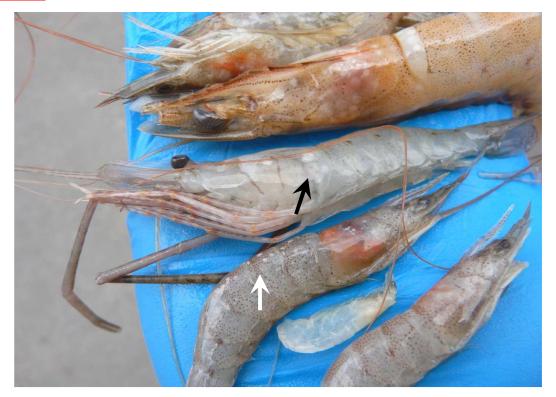


Figure 5. Signs of clinical WSD on locally caught *Metapenaeus* (white arrows) and *Macrobrachium* sp (black arrows) being sold as bait on the morning of 11th December. Note: testing of these samples gave indeterminate results (WSSV positive for some tests, but not others)



Figure 6. Signs of clinical WSD on locally caught Palaeomonid shrimp being sold as bait on the morning of 11th December.

Note: testing of these samples gave indeterminate results (WSSV positive for some tests, but not others)

- 7. Upon informing the tackleshop staff of the finding and showing them the specimens, they agreed the prawns looked to have WSD based on educational materials provided to them by Biosecurity QLD staff the previous day. They then immediately agreed to remove the prawns from sale, I personally instructed some customers not to buy the infected prawns (they were scooping them up to buy them) and instead I purchased around \$55 worth of bait prawns to remove them from sale and collect them as samples for later DNA analysis. I instructed staff to place the samples in their freezer on the premises then notified Biosecurity QLD staff that I reasonably suspected sale of WSD affected bait prawns in the movement control area. I then removed gloves and washed hands with alcohol handwash to decontaminate them before continuing on to 3 IP.
- 8. Upon entering 3 IP around 12.30 pm and undertaking routine decontamination I began to sample prawns from ponds 1 and 5 for fixation into 95% ethanol, 5% methanol. Samples of 60 were randomly collected from each pond using a castnet, examined, the head was then removed with scissors and placed in fixative. Prawns from both ponds looked healthy. The sampling was interrupted by a need to collect more fixative, which necessitated a run to the local hardware for methylated spirits. Upon returning I dropped into Gem Bait and Tackle again to check up that the bait prawns had been removed from sale and caught up with 2 Biosecurity QLD officers, explained what happened earlier that day and showed them the suspected WSD infected prawns, including the Palaeomonid shrimp in Figure 6. The staff showed the officers their freezer with stockpiled locally caught prawns, some of which may have been weeks to months old these would appear to be perfect samples for tracing the epidemiology of the epizootic in wild prawns in the Logan

River and the officers intended to seize the entire stock. The officers also seized my samples, and I discussed the risks posed by sale of yabbies (*Callianassa* spp.) which were held live in trays (Figure 7) on the premise near suspected WSD infected crustaceans (where cross contamination by staff handling fresh bait prawns would be almost certain). I strongly recommended that the yabbies should also be seized, removed from sale and destroyed and that the entire aquarium setup should be chlorinated and dried out.



Figure 7. Live yabbies (*Callianassa* spp.) being sold as bait from a premise with large quantities of locally caught, suspected WSD infected prawns. The chances of cross contamination of yabbies by staff was considered to be extremely high, such that I recommend these should be seized and destroyed and the system holding them chlorinated and dried out.

- 9. After leaving the tackle shop I returned to finish sampling prawns from pond 1 at 3 IP but as I finished at 4 pm I was interrupted by a phone call from one of the Biosecurity officers asking me to return to the tackle shop to talk to an aggrieved river beam trawl operator and tackle shop owner who were asking why the whole shipment of prawns needed to be seized. I explained the extreme risk of spreading the disease via bait and how co-operation with authorities was the best option under these circumstances. The trawler operator then understood the situation better and offered an observation that he noticed something unusual regarding banana prawns that had large catches in early May 2016, but suddenly dropping to nothing literally overnight, co-inciding with a sudden drop in water temperatures around that time in Moreton Bay (B.K. Diggles, unpublished data). The trawler operator suggested that in the previous 20-25 years he had been operating, he had never observed a similar event, hence forensic examination of the bait prawns seized at Gem Tackle would appear very useful to try to ascertain how long WSSV has been present in the river (possibly since May 2016?). I left to return and clean up equipment and store samples at 3 IP at 4.30 pm.
- 10. After completing tasks at 3 IP and routine between farm decontamination (decontaminating footwear, camera, phone, hand washing and replacement of boots, outer clothing etc which I do before entering each site), I moved on to conduct an initial site inspection at 4 IP, arriving there at 5 pm. Here Elwyn Truloff confirmed that the initial infections were seen in ponds 1 and 3 and that clinical disease was noticed just prior to chlorine treatment on 9th December. I walked the entire farm noting that there were very high numbers of jelly prawns mixed in with *P. monodon* in ponds 1 and 3. Elwyn confirmed that ponds 1, 2, 3, 4 were chlorinated 2 days ago, ponds 5 and 6 were chlorinated yesterday, and that ponds 7, 8 and 24 were chlorinated today as evidenced by the fresh bodies of prawns in those latter 2 ponds.
- 11. At 6.44 pm I noted a pair of grapsid crabs still feeding on dead prawns in chlorinated pond 5, over 24 hours after treatment. Both crabs were on the dirt at the pond edge, but both reentered the chlorinated water upon my approach. I saw no evidence of clinical disease in any ponds but observed some unusual behaviour in ponds 17 and 18 which I will check up on again tomorrow. I decontaminated and left the farm at 7.15 pm to return to my accommodation.

# Appendix 5. WSD outbreak situation report #5. 13 Dec 2016

This is a summary of developments along the Logan River since my last report of 11<sup>th</sup> December 2016.

### 12<sup>th</sup> December 2016

1. On Monday 12<sup>th</sup> December I began by collecting Australian frozen bait prawns from fishing tackle shops outside the control area in Southport in order to archive some samples which could be tested for WSSV at a later date. I then visited shops in the control area. Proprietors at one tackle shop at Cabbage Tree Point informed me that Biosecurity QLD staff had visited the previous day and seized locally caught prawns that were on sale. However other tackle stores at Cabbage Tree Point and Jacobs Well were still selling packets of bait prawns, albeit brands that appear to have come from locations outside the control zone, such as Brisbane River (Castaway Brand) and Tweed Bait.

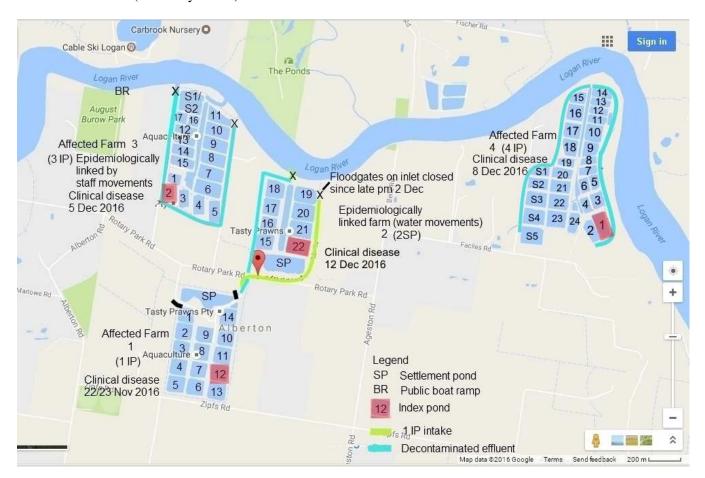


Figure 1. Locations of farms, including pond numbers and location of index ponds at each site.

2. While I was at Jacobs Well I was contacted on the phone by the proprietor of Gem Bait and Tackle who informed me that he had bought in some new stock of bait prawns sourced from outside the control area, but that some of those prawns appeared to have white spots on them. I advised him that if there was any doubt, do not sell them to the public and notify Biosecurity QLD if he suspected whitespot disease in any bait prawns. I was also asked about how to decontaminate the 800 litre aquarium system there that was holding the live yabbies. I calculated a dose equivalent to 100 mg/L chlorine based on volume and provided him with advice that it was probably better that Biosecurity QLD administered the chlorine and

monitored efficacy and chlorine availability in the trays (especially as subsequently it was revealed that a biofilter in the system would need to be decontaminated as well).

- 3. After archiving samples of bait prawns on ice and decontamination (hand wash) I proceeded to 3 IP where I arrived at 11 am and entered the decontamination checkpoint, notified the site controller of my presence and passed through the decontamination area (gumboots, footbath, handwash). There I was informed by Biosecurity QLD staff that ponds 6 and 7 had tested positive for WSSV by PCR and that they were to be decontaminated ASAP. I took 3 new castnets from the storage shed (purchased earlier in the week) and took random samples of prawns from ponds 6, 7 and 10 for fixation and archiving on site in 95% ethanol. There was no obvious mortality or abnormal behaviour in prawns from any of the ponds, however at least 4 of the 60 prawns sampled from pond 7 showed clinical signs of WSD including white spots and reddening of the carapace. I also observed a large number of jelly prawns (*Acetes* sp.) in ponds 6 and 7. I completed sampling of pond 7 just prior to the chlorine trucks arriving with chlorination of pond 7 starting at 1.45 pm.
- 4. After completing decontamination of hands and equipment (ethanol and detergent washing and scrubbing) I began sampling of pond 6 (with none of the 60 prawns showing any clinical signs of WSD), finishing by 2.45 pm. During the process of storing the sample on site and decontamination of hands and equipment, I observed the chlorine being administered to pond 6 by 3.10 pm. I then moved to sample pond 10 but briefly stopped to observe the chlorinated prawns in pond 7. Both *P. monodon* and jelly prawns were swimming in an agitated state on the water surface and I estimated that around 7-10% of the *P. monodon* that were visible were showing clinical signs of WSD with visible white spots on the carapace.
- 5. Moving to pond 10 I observed Biosecurity QLD staff castnet sampling prawns from ponds 9 and 10. Their methodology included sanitation of castnets in virkon for 5 minutes or so between ponds, slightly different to my approach of using a new castnet per pond. I started my own sampling of pond 10 around 3.20 pm and completed sampling and fixing 60 prawns from that pond by 4.15 pm. None of the prawns from pond 10 showed any clinical signs of WSD. After decontamination of hands and equipment (scrubbing in ethanol and detergents) and storage of the ethanol fixed samples on site I observed the experimental emergency harvested, cooked prawns being taken on ice in sealed fully enclosed containers to freezers nearby (presumably at 1 IP) for archiving and storage.
- 6. I then proceeded to decontaminate boots and hands at the farm gate before changing to new clothes and gumboots before proceeding to 4 IP. My conversations with Elwyn at 4 IP disclosed that a small number of prawn mortalities observed in pond 3 at that site may have been due to lightning strike, meaning that the true index case for that farm was pond 1 where clinical signs of disease were first observed on the evening of 8 December 2016 (Figure 1). I was informed by Biosecurity QLD staff who I reported to on site that decontamination of ponds 20, 21, 22, 13 and 14 had been undertaken at 4 IP today.
- 7. I took 2 new castnets purchased that day and took random samples of 60 prawns from ponds 8 and 18 for fixation and archiving on site in 95% ethanol. Several prawns from pond 8 had what the farmer referred to as "redback" syndrome with focal bilaterally symmetrical reddish areas along the dorsal cephalothorax and abdomen of affected prawns. A low number of prawns also had "saddleback" like deformities of the abdominal segment just before the telson. Other than these, there was no obvious lesions, mortality or abnormal behaviour in prawns from either pond. The used castnets were packaged up and left at the side of the sampled ponds.
- 8. After decontaminating hands and equipment and just prior to leaving the property Elwyn Truloff was provided with hard copy documents for the first time that outlined the results of testing (from AAHL and Biosecurity QLD) and other information explaining what WSD is and the significance of it to Australia (extracts out of the Aquatic Animal Diseases Significant to Australia Field Guide (4th Ed), Biosecurity QLD information leaflets, etc.). Upon his request, I went through the documents with him explaining various aspects. It was noted that provision of this information in a written form much earlier in the response would have been desirable as it would have allayed much confusion and stress at the farm people at the coalface are often the last to hear of developments in the response process and therefore regular

communication and updates (in written form as this provides substance) can assist in making response processes more transparent (thus improving compliance and reducing stress for those most affected at the coalface).

9. I decontaminated and left the farm at 7.45 pm to return to my accommodation. Driving home I was informed by Ian Rossman that prawns in pond 22 at 2 SP had begun to show signs of disease that morning (am of 12 Dec 2016) and asked whether I could check them out tomorrow.

## 13<sup>th</sup> December 2016

- 10. At 7.45 am I was contacted by Rob King, environmental officer of Sunfish QLD, to inform me that he and some other Sunfish members had observed bait prawns with apparent white spots on them sometime in the recent past, possibly approximately 2 months previously. I informed him to try to obtain photos, samples or some other evidence which could be acted upon.
- 11. At their request I briefly stopped at Gem Bait and Tackle at 8.45 am to observe their progress with decontaminating the yabby aquarium system and look at the suspect batch of bait prawns they had bought in from outside the control area yesterday. The trays of the yabby aquarium system did not smell of chlorine (probably neutralised by the biofilter) so I informed manager Bruce that he should seek help from Biosecurity QLD to decontaminate the system properly. Bruce then took me to the fridge at the back of the shop where a container of thawed prawns was held with a sign on them saying "not for sale". I got Bruce to take them out into the sunlight and noted that some of the prawns (*Metapenaeus* sp.) had large numbers of small white spots on the carapace (Figure 2). While I was not certain the signs were necessarily indicative of WSD and could instead represent a storage/processing artifact, under the circumstances their decision to withdraw them from sale appeared the correct one, and I rang Steve Wesche to inform him of the sample and requested that Biosecurity QLD officers attend the site ASAP to collect the prawns and decontaminate the area.
- 12. After decontaminating hands and putting on gumboots I entered the decontamination checkpoint at 3 IP, notified the site controller of my presence, signed in and entered the cookout building to change the fixative for one of the sample jars stored on site (held in the fridge near their cooking facilities). I then exited 3 IP (sign out, footbaths, hand wash) changed gumboots and parked outside 2 SP at 9.45 am where I was met by Ian Rossman and the Biosecurity QLD site controller. We proceeded on foot to pond 22 where I observed clinically affected prawns with WSD swimming on the surface of the pond and also moribund in the shallows (Figure 3). There were also dead prawns every 1 to 2 meters along the pond edge. There was only 1 bird control person on site at the time and as it was clear that the pond had WSD and he could not leave it unattended to patrol the remainder of the site, I asked him to call in extra bird control personnel and began to sample 60 prawns for fixation into ethanol using a new castnet under supervision of the site controller. It must be noted that pond 22 was also the pond furtherest away from the intake pumps on 2 SP (Figure 1) hence it was, like all other infected farms, taking water from the end of the supply channel. This consistent picture throughout all 4 farms infected so far suggests the spread of the disease between farms is not random or likely to be caused by movements of birds or human activities, but instead is highly likely to be associated with pumping in virus (or more likely, hosts infected with the virus) from the river.



Figure 2. Suspect thawed prawns obtained from outside the control area that were withheld from sale at Gem Bait and Tackle on 13 December 2016. While the lesions (arrow) are not round spots and may represent a storage/processing artifact, they should be PCR tested to rule out WSD.



Figure 3. Clinically diseased moribund prawn at the edge of pond 22 on 2 SP at 10 am 13th Dec.

- 13. After sampling 60 prawns from pond 22, of which 7 had clinical signs of WSD, I packaged up the castnet and left it at the pond side, decontaminated hands and equipment (ethanol, scrubbing with detergent) and at 11.30 am looked for the site controller to ask him where the samples could be stored. However the site controller had left the farm and could not be found, hence I informed the two bird control personnel (Daniel Franey and David Willoughby) that the fixed samples would be stored in the feed shed (on the ground to the left hand side of the southern door). I then decontaminated hands and gumboots (footbath, hand sprays) and all equipment at the farm gate then changed clothes and waited in my car outside the gate for the site controller to return. After having lunch I waited until 1 pm at which time the site controller had still not returned, so I then moved on to 4 IP.
- 14. Arriving at site 4 IP I left the car outside the front gate and donned a third pair of gumboots then notified the site controller (Damien Bougoures) of my intent to sample 3 ponds for fixation of prawns into ethanol to be stored on site. I was informed that all of the discharge drains had now been decontaminated with chlorine (Figure 1) and that Ponds S1 and S2 had also been treated. I was then taken down to the ponds by Elwyn Truloff with 3 castnets and randomly sampled 60 prawns into ethanol fixative from ponds 10, 11 and 17 between 1.15 pm and 6 pm.
- 15. I saw no evidence of clinical disease in any ponds visited on 4 IP today except for some tail deformities in ponds 10 and 11. I packaged up the castnets, left them at the pond side, decontaminated equipment (ethanol, detergent scrub) and hands, stored the samples on site in a fridge in the main harvesting building, and after assisting Elwyn to fix his electric farm gate (the entry key switch had been damaged by some of the site visitors), I left the farm at 6.25 pm to return to my accommodation.

# Appendix 6. WSD outbreak situation report #6. 30 Dec 2016

This is a summary of developments along the Logan River since my last report of 13<sup>th</sup> December 2016. The current situation with respect to infected premises can be found in Figure 1 (over page).

## 14<sup>th</sup> December 2016

1. On Wednesday 14<sup>th</sup> December after the industry phone conference (where it was mentioned chlorination of ponds on 3IP was scheduled for completion that day), I inspected several supermarkets in the vicinity of Southport and witnessed thawed frozen raw prawns from various countries (China, Thailand, Malaysia) being sold at multiple Woolworths and Coles outlets by the kg over the counter without any signage or advice to consumers not to use them as bait.

### 15th December 2016

- 2. On Thursday 15<sup>th</sup> December after the industry phone conference call I participated in a National Industry wide phone conference with State and Federal Biosecurity authorities where official updates for the WSD outbreak situation were given. One very important piece of information that was provided by Federal authorities was that as of May 2016, of the 448 consignments of imported frozen raw prawns bought into Australia, 73 consignments (16.3%) tested positive for whitespot virus. It will be important to ascertain the exact sampling methodologies used by Federal biosecurity and border security staff to arrive at these figures, so that the potential likelihood of false negative test results in any non-positive consignments can be better assessed.
- 3. At 4 pm I inspected Gem Bait and Tackle and witnessed that all the freezers which had contained frozen prawns had been shut down, cleaned out and chlorinated by Biosecurity QLD (Figure 2), as had all of the recirculation systems that housed live yabbies. Staff were in the process of restocking the freezers with bait prawns sourced from outside the fishing closure area.
- 4. At 4.20 pm I inspected the Alberton Boat ramp and found that there were no advisory signs informing boaters or fishers of the fishing closure conditions. I spoke to the farmers at 3IP and they confirmed that all ponds and drains had been chlorinated including settlement ponds. I phoned the farmers at 4IP and they confirmed that the last 5 ponds on their property had been chlorinated today. I then parked outside 2IP where there was still no supervision or signage informing traffic along Rotary Park Rd not to fish in the 1IP intake. Not being able to locate a Biosecurity QLD site controller, I entered the decontamination area, then spoke to the single bird control officer on site, who was placed near the index Pond 22 (Figure 1), which was chlorinated late pm on 14<sup>th</sup> December. There were large numbers of dead *P. monodon* and jelly prawns visible along the edges and pond margins (Figure 3). I then inspected the remaining ponds 21, 20, 19, 18, 16 and 15, then sampled 60 prawns from Pond 17 for gentics and left the samples fixed in 95% ethanol inside the feed hut at 7 pm, before decontaminating and returning home.

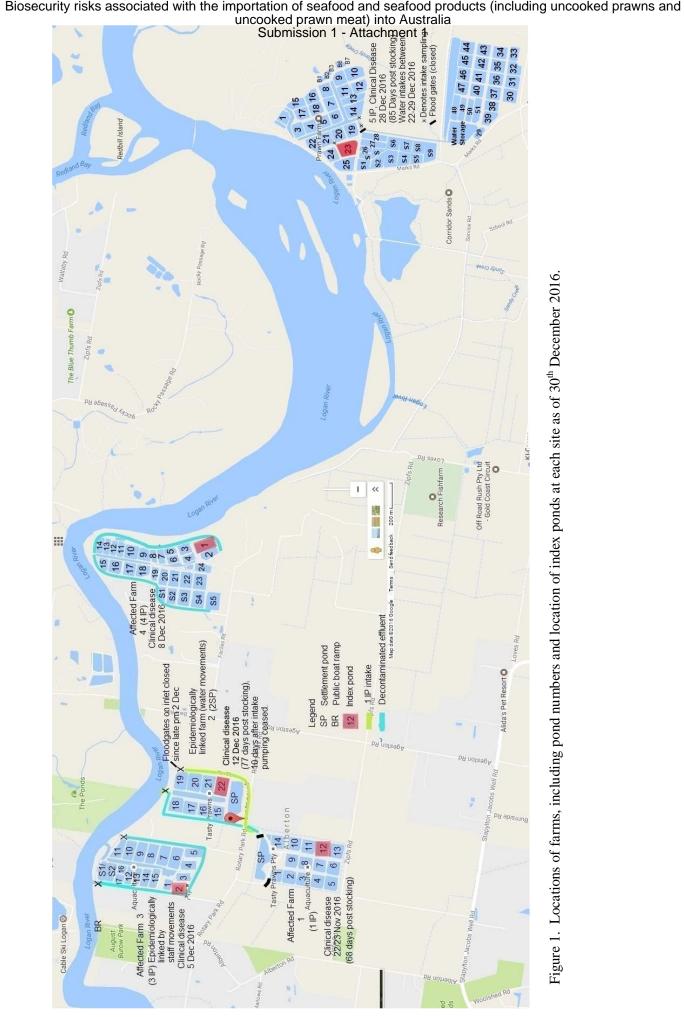


Figure 1. Locations of farms, including pond numbers and location of index ponds at each site as of 30th December 2016.



Figure 2. Cleaned and chlorinated freezers at Gem Bait and Tackle on 15 December 2016.



Figure 3. Large numbers of dead *P. monodon* and jelly prawns were visible and accessible to birds along the edges of pond 22 at 24 hours post chlorination on 15 December 2015. Only one bird control officer was on site that evening.

### 16<sup>th</sup> December 2016

5. On Friday 16<sup>th</sup> December I was phoned by Dan from 3IP at 9 am who informed me that he had just witnessed a live prawn swim over to him at their pond 1 that morning, 3 days after it had been chlorinated. He thought it may have survived by hiding near the inlet boards where a dilution effect may have occurred. On the industry phone conference at 9.30 am we were informed that decontamination of all drains and inlets of 3 IP and 4 IP was to be completed today and that 2 IP would be completed tomorrow. A phone hookup with the Biosecurity QLD technical working group at 11 am disclosed that some of the earlier bait samples I had obtained from Gem Bait and Tackle tested positive, others were borderline positive /indeterminate, but the species of the positive sample (Banana prawn, *Metapenaeus*, Palaemonid) was not certain/known.

## 17-20<sup>th</sup> December 2016

6. From Saturday 17<sup>th</sup> till Tuesday 20<sup>th</sup> December I participated in various phone conferences (Industry, Biosecurity QLD, Disposal and Decontamination Technical Working Group). Notes from these conferences are available if required.

### 21st December 2016

- 7. On Wednesday 21<sup>st</sup> December I participated in an industry phone conference and a technical working group phone conference. In the Technical Working Group phone conference it was disclosed that the samples from Gem Bait and Tackle which I had identified and fixed representative samples in 95% ethanol prior to them being seized on 11<sup>th</sup> December were PCR positive, but the species that were positive was not identified. It was disclosed, however, that "3 or 4" of the bait prawns sampled had real time PCR CT values of 32-35, but histology showed no confirmed inclusions. It must be noted that histopathology of bait samples that had been frozen for possibly 2 or more weeks prior to fixation in 95% ethanol would result in many artefactual changes that may greatly reduce the sensitivity of histopathology as a diagnostic tool under such circumstances, especially with prawns due to the fact that crustacean tissues autolyse very quickly after death.
- 8. In the PM, I briefly inspected 2IP at 2.30 pm with the then site manager (Perry Jones) to ascertain that chlorination of all ponds there had been completed, and witnessed that chlorination of the settlement pond there was about to commence. I then attended a meeting at 1IP with Biosecurity QLD and the affected farmers from 1, 2, 3, and 4IP, where strategies for finalizing decontamination and beginning the process of draining and disposal were discussed. The meeting finished with resolutions from Biosecurity QLD to finalise disposal strategies in close collaboration with farmers.

## 22-28<sup>th</sup> December 2016

Dec) where it was disclosed that the South Australian Government had prohibited entry of live or dead (uncooked) crustacean of the order Decapoda from the Logan River area. Then at 8 pm on the 28<sup>th</sup> of December Helen Jenkins rang to inform me that there were signs of unusual behaviour in prawns from Pond 23 at 5 ARP, that samples had been sent off for testing, and to stand by for more information. The last time the pond had been sampled for testing was Thursday 22<sup>nd</sup> December, which apparently gave negative results. However since that time the farm had taken in approximately 54,450 tonnes of water at the top of the tide since 22<sup>nd</sup> December (approx 1.5 tonnes per second for the following time periods 22 Dec – 120 min between 3.40-5.40 am, 23 Dec- 300 min between 4.20 to 6.50 am and 14.20 till 6.50 pm, 24 Dec – 95 min between 5.35 am and 7.10 am, 29 Dec- 90 min between 11 am and 12.30 pm) that was not treated.

### 29th December 2016

8a. On Thursday 29<sup>th</sup> December Helen Jenkins rang to confirm the samples from Pond 23 on 5 ARP were positive by PCR for WSSV. Hence 5 ARP was now designated 5IP. After a brief phone conversation with Nick Moore at 9.25 am (who was trying to keep terns away from infected pond 23 just as bird mitigation arrived) I organized to visit 5IP on 30 Dec. It was noted by Nick Moore that use of non-lethal ammunition simply moved birds that were feeding on pond 23 away to other ponds on site and that distribution of diseased prawns from pond 23 to other ponds within 5IP (including pond 25) was observed by him.

### 30<sup>th</sup> December 2016

- 9. On Friday 30<sup>th</sup> December I was rung by Bomber Lancaster (5IP site controller) and briefed that I should liase with Brian Paterson on farm for any sample collection etc. that was to be performed that day. I arrived on farm at 8.30 am to see chlorine trucks adjacent to ponds 23 and 25 (Figure 1). Pond 23 had been chlorinated with 24 tonnes of chlorine the previous evening and Pond 25 was emergency harvested early am that morning and was partially drained (Figure 4). After topping up chlorine levels in pond 23 with an additional 15 tonnes of chlorine, the truck put the remaining chlorine (5 tonnes?) into pond 25 as the level was being raised back up with water from the adjacent effluent canal. I was informed by Nick Moore and Alistair that the adjacent ponds 26, 27 and 28 (Figure 1) had been harvested earlier (date to be confirmed) with 98-100% survival. The survival data for prawns from Pond 25 that was harvested that morning was not available at the time, however a small number of clinically diseased prawns were apparently noted by 5 IP staff from that pond.
- 10. During discussions with Brian Paterson he mentioned seeing sluggish prawns at ponds 21, 20, and 19. I inspected these ponds and confirmed that moribund prawns with clinical signs of WSSV were visible along the pond edges of ponds 21 and 19. A group of half a dozen seagulls were roosting between ponds 22 and 21 so I informed Biosecurity QLD staff that pond 21 was also infected and that bird control needed to look in that direction as well. There were more birds on 5IP than at previous farms including a flock of approximately 60 seagulls between ponds 15 and 16 (Figure 5), and bird control staff were observed to be using a mixture of both non-lethal and lethal ammunition. A delivery of 6 new plankton nets was delivered to the farm gate around 9.30 am and I worked with on site fabricator Sean to rig them up with poles so that plankton could be sampled from the intake canals.
- 11. Fabrication of the plankton net poles was completed by 1.30 pm (Figure 6) and during the morning and afternoon I had several meetings with Biosecurity QLD staff, Nick Moore, Alistair Dick and Noel Herbst to get up to speed with the farm layout and their harvesting plans. Moving back onto the farm at 1.30 pm I noted that the next chlorine truck was already treating pond 21, however chlorine availability or equipment breakdown prevented treatment of ponds 20 and 19.
- 12. In then went to inspect the ponds on the eastern side of the farm and noted that ponds 1, 15, 17, 16, 18, 8 and 9 were all lined (as are most/all the ponds in this section of the farm) and looked normal. I did note one sluggish prawn along the eastern edge of pond 11 at around 4.30 pm. I then proceeded to the edge of pond 19 under supervision of Brian Paterson to collect plankton samples at 3 sites (marked x in Figure 1) between 4.48 pm and 5.20 pm (Figure 7).



Figure 4. Chlorination of pond 25 (left) with leftover chlorine from index pond 23 (right) at 5IP at 9 am on 30<sup>th</sup> Dec. Pond 25 is partially drained from the emergency harvest that morning.



Figure 5. More birds are present on 5IP than other farms I have inspected. Here a flock of 60 seagulls roost between ponds 15 and 16.

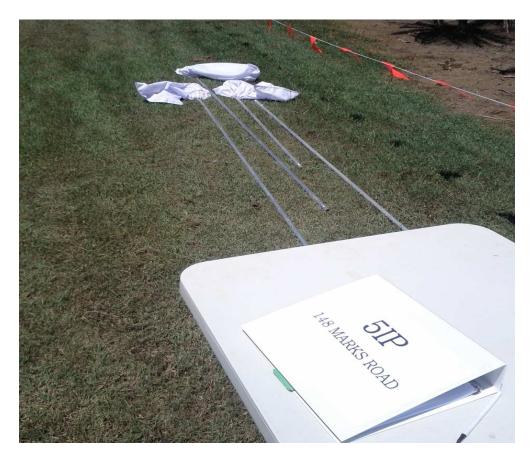


Figure 6. Plankton sampling nets rigged and ready to go courtesy of fabricator Sean.



Figure 7. Brian Paterson taking plankton samples at the southern end of the 5 IP intake near pond 19.

- 13. Keen to observe what was happening in the intake channels at night, I returned to the same 3 plankton sampling locations after 7 pm in the dark to repeat the sampling process. When resampling the main intake channel, I noticed a vast increase in crustacean activity compared to 2 hours previous. Indeed in the torchlight I was surprised by the amount of visible crustacean life (mainly large numbers of banana prawns\*) at the very end (rather than down the sides) of the intake channel. I managed to capture and inspect 10 to 12 of the banana prawns (which were apparently healthy) with the plankton net (using it as a scoop net) and fixed them in ethanol for later analysis, but I could have sampled 60 to 100 or more if I had a castnet. I intend to return tonight and resample the intake properly (hopefully with Biosecurity QLD supervision) with a new castnet to quantify the number of animals along the sides of the intake vs at the ends, because I estimate 5 to 10 times more animals at the ends of the intake compared to along the sides.
- 14. After completing plankton sampling and fixing samples in 50% ethanol, I stored the plankton samples and banana prawn\* samples on site decontaminated boost and equipment (all left on site with the samples) and left at 9 pm. I intend to return today (on the 31<sup>st</sup>) to examine the plankton samples under the microscope to see if any crustacea were actually collected with the plankton net (unsure at present), and am liasing with the site controller to organise Biosecurity QLD staff to be present to take samples from the intake at night.

These were later identified as Eastern king prawns *Penaeus plejebus* – see situation report #7

# Appendix 7. WSD outbreak situation report #7. 31 Dec 2016

This is a summary of developments along the Logan River since my last report of 30<sup>th</sup> December 2016. The current situation with respect to infected premises can be found in Figure 1 (over page).

#### 31st December 2016

- 1. On Saturday 31<sup>st</sup> December after the industry phone conference, I had conversations regarding surveillance with Mark Cozens from the CCEAD, before writing industry situation report 6. Then departed for 5IP arriving at 3.45 pm. I checked in with site controlled Vern who informed me that ponds 19, 20, 22 and 24 were chlorinated today (Figure 1). I informed him of my intention to go to the laboratory to assess the plankton samples taken yesterday to see if there were in fact any plankton in them. I also informed him of my plans to conduct a night time cast net survey at various positions along the sides and ends of the main intake channel to ascertain whether the distribution of prawns and/or other crustaceans was uniform or biased towards the ends of the channel (as was observed the previous night).
- 2. I caught up with Alistair who kindly showed me to the laboratory and organized a dissecting and compound microscope to examine the ethanol fixed plankton samples collected the previous night from the sites shown in Figure 2. Investigation of the mesh size of the net used under the microscope suggests it was a phytoplankton net of approx 15-20  $\mu$ m mesh size, which is too small to properly sample zooplankton with confidence (several zooplankton nets with 100 $\mu$ m mesh size have since been ordered). Inspection of all the plankton samples was not possible in the time available, however I did manage to assess 3 of the 6 samples and enumerate 2 of them (Table 1).

Table 1. Details of plankton samples taken with a  $15\text{-}20\mu\text{m}$  mesh net on 30 Dec 2016 in the 5IP intake. The samples taken after dark at site A had 13 times more plankton in them as well as higher species diversity than the daytime samples. TBA = results to be announced.

Sample	Time	Location	Result
name	taken		
A	4.48 pm	Southwest branch of intake near	N = 3 including
		pond 19	1 Lepostraca-like copepod
			2 harpactacoid copepods
В	5.08 pm	Southern end of main intake	TBA
		channel	
C	5.20 pm	Intake canal to ponds 23, 24	TBA
D	7.00 pm	Southwest branch of intake near	N = 41, including 23 insect larvae,
		pond 19	10 harpactacoid copepods
			1 cyclopoid copepod sp. 1
			5 Cyclopoid (?) copepod sp. 2
			2 Acetes sp. (jelly prawn) larvae
E	7.45 pm	Southern end of main intake	High diversity and numbers of plankton,
		channel	including fish (mullet) larvae, Acetes (jelly
			prawn) larvae, Calanoid copepods, Mysids (?),
			zoea, etc. Full results TBA.
F	8.10 pm	Intake canal to ponds 23, 24	TBA

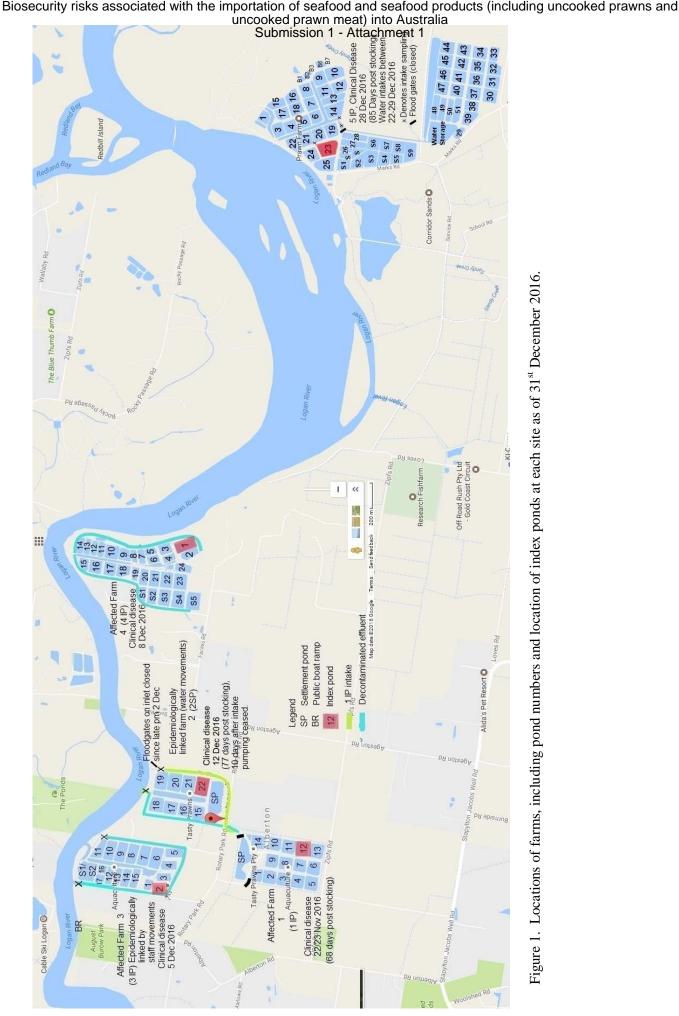


Figure 1. Locations of farms, including pond numbers and location of index ponds at each site as of 31st December 2016.

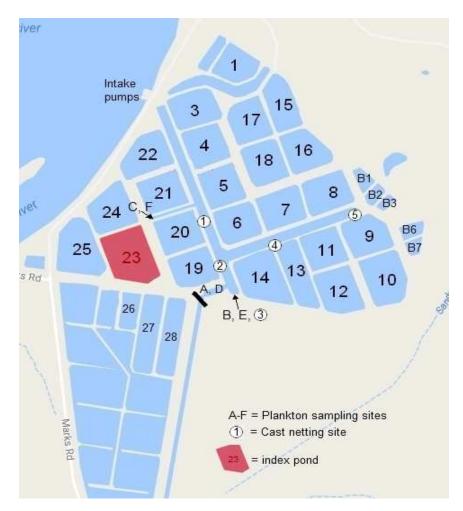


Figure 2. Locations of plankton and cast net sampling sites at 5IP.

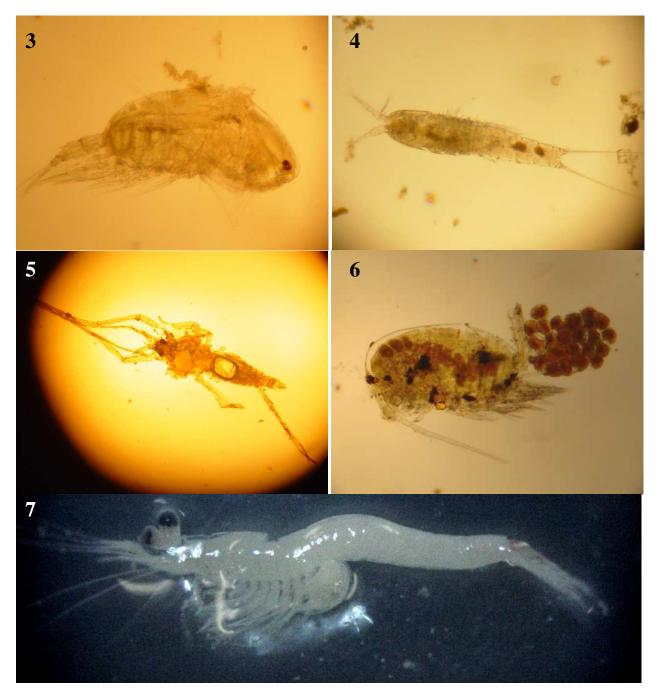
Table 2. Timing and results of cast net sampling from sites 1-5 between 7.30 and 10 pm of 31 Dec 2016. Results are sum totals from 5 random casts after at least one *P. plejebus* was visible on the surface.

Site	Time taken	Location	Penaeus	<u>Penaeus</u>	Metapenaeus	Total	% of total
			plejebus	<mark>monodon</mark> *		catch	catch
1	7.30 pm	East side of pond 20	15	3	1	19	10.4%
2	8.00 pm	East side of pond 19	30	4	12	46	25.3%
3	8.30 pm	Southern end of main	41	4	7	52	28.6%
		intake channel					
4	9.00 pm	North intersection of	8	0	5	13	7.1%
		ponds 13, 14					
5	9.30 pm	Eastern end of east	39	7	6	52	28.6%
		intake channel					
Total	10.00 pm	All sites	133	18	31	182	100 %

#### Note: identification subsequently revised to *Penaeus* spp., see Situation report #9.

3. The main feature of the plankton samples examined was the marked difference in species diversity and numbers of plankton sampled between daytime and nighttime. The samples taken from the southwest branch of the intake near pond 19 (Samples A and D) had 13 times more plankton and over double the species diversity when sampled at 7 pm (sample D) than during daylight 2 hours previously (Sample A). Daylight samples comprised a total of 3 Lepostraca-like (Figure 3) and harpactacoid (Figure 4) copepods

while at night the samples were numerically dominated by insect larvae (Figure 5), and various other crustaceans species including cyclopoid-like copepods (Figure 6) and larval jelly prawns (*Acetes* sp., Figure 7). All of these groups are known vectors for WSSV and representative samples remain fixed in ethanol on site for later PCR analysis if required. Prior to decontamination of the intake channels it would be extremely valuable to not only enumerate the small number of plankton samples currently available prior to PCR analysis, but also significantly increase the numbers of samples taken at various locations with a zooplankton net of proper mesh size in order to better understand the distribution of zooplankton in the intake channel and obtain more zooplankton samples for WSSV testing.



Figures 3-7. Various types of plankton collected from the intake channels at 5IP. All are known hosts or vectors of WSSV. 3. Lepostraca-like copepod, 4. harpactacoid copepod, 5. insect larvae, 6. Cyclopoid-like copepod, 7. larval jelly prawn (*Acetes* sp).

- 4. After cleaning up the laboratory and storing the plankton samples in 70% ethanol, I alerted bird control officers of my presence and with their escort entered the farm with a new castnet to sample the intake channel at 5 sites (sites 1-5, see Figure 2, Table 2). Wind strength was from the north west estimated at 15 to 20 knots. Each site was sampled using 5 casts of the net, with the timing of each throw coinciding with torchlight spotting of at least one eastern king prawn (*Penaeus plejebus*, ID now confirmed after mistakenly referring to them as banana prawns in the previous report). The results of the cast net sampling are summarized in Table 2. There did not appear to be as much *P. plejebus* activity near the southern end of the main intake channel (site 3), as there was on the night of 30<sup>th</sup> December, nevertheless sampling the ends of both intake channels resulted in the highest number of prawns (n = 52 from both sites 3 and 5), most of which were fixed in 95% ethanol for later analysis. In the main north-south channel, crustacean (and fish) activity was concentrated at the downwind sites 2 and 3, both of which had over twice the number of animals sampled at site 1, while in the east-west channel, there were 4 times the number of crustaceans sampled at the end of the channel (site 5) compared to the side of the channel (site 4).
- 5. While the main species sampled was *P. plejebus* (Table 2), most of which appeared healthy and active (though a few had melanised lesions on the telson or abdominal segments these individuals were all fixed in ethanol), a smaller number of *Metapenaeus* and *Penaeus monodon* were also sampled (Figure 8). Some of the latter two species appeared to be sluggish with soft shells, hence all *P. monodon* and *Metapenaeus* were fixed in ethanol for later analysis. Other incidental captures included 4 striped trumpeter (*Pelates* sp., one each from sites 1, 3, 4, 5), 3 fanbellied leatherjackets (*Monacanthus chinensis*, two from site 1, and one from site 2), 3 striped anglerfish (*Antennarius striatus*, one each from sites 1, 2, and 3), one toadfish (*Tetractenos hamiltoni*, from site 1) and 3 squid (from site 5).



Figure 8. While the majority of prawns sampled from the 5IP intake were identified as eastern king prawns (*Penaeus plejebus*, top), a smaller number of *Penaeus monodon* (below) and greasyback prawns (*Metapenaeus*, not pictured) were also sampled. \*Note: identification subsequently revised to *Penaeus* spp., see Situation report #9.

6. After completing cast net sampling and fixing samples in 95% ethanol, decontaminated boots and equipment, then stored the samples on site in the laboratory fridge, and left the farm at 10.30 pm.

# Appendix 8. WSD outbreak situation report #8. 9 Jan 2017

This is a summary of developments along the Logan River since my last report of 31<sup>st</sup> December 2016. The current situation with respect to infected premises can be found in Figure 1 (over page).

## 1<sup>st</sup> January 2017

1. On Sunday 1<sup>st</sup> January 2017 I spent 12 hours report writing (Sitrep report #7) and in phone communications with Helen Jenkins and Nick Moore. Helen informed me that Biosecurity OLD had reported a WSSV positive sample in a single mud crab (Scylla serrata) sampled on 23<sup>rd</sup> December next to the road in the outlet canal down from Rocky Point Farm (7ARP) (Figure 2). The crab was lightly infected based on a real time (quantitative) PCR CT value of 40.46, however the result was confirmed as reliable by repeat testing with a 10x dilution of the sample returning a CT value of 44.98. The CT value is a quantitative measurement of the amount of viral DNA present in the original sample. Real time PCR (also known as quantitative PCR or qPCR) monitors the amount of target DNA that is amplified during each PCR cycle (i.e. in real time during the PCR process, not only at the end as in conventional PCR). The CT value is a measure of the number of PCR cycles required to exceed a certain predetermined threshold amount of target DNA (i.e. cycle threshold value or CT value). There is an inverse relationship between viral load and CT number because the threshold is reached in fewer PCR cycles when there is more viral DNA in the original sample, i.e. a high amount of virus gives a low CT value. To put the mudcrab result in context, a CT value of 40 is relatively high compared to the usual positive threshold CT levels chosen by some laboratories of 30-32, indicating the original sample from the mudcrab had a relatively low number of viral copies per unit weight of tissue, possibly representing a carrier status.

# 2<sup>nd</sup> January 2017

- 2. On Monday 2<sup>nd</sup> January the morning was spent ordering plankton nets and in communications with Serena Zipf who informed me that Biosecurity QLD would be treating the infected section of the 7ARP outlet canal and taking more samples from her ponds at 7ARP, which remained negative. They would not be allowed to harvest any prawns until the results of the latest tests became available. I left home at 11.30 am and purchased additional new cast nets and ethanol fixatives, before arriving at 5IP at 3.45 pm. There I checked with site controller Vern who confirmed that ponds 43, 44 and 45 had been chlorinated that day (Figure 1). I informed him of my intention to go to the laboratory to assess the remaining plankton samples taken 2 days prior. I also informed him of my plans to assist Biosecurity QLD field collection staff (Simon and Daniel) to collect night time plankton samples and cast net samples at various positions along the sides and ends of the main intake channel.
- 3. Inspection of the remaining 3 plankton samples taken with a 15-20µm mesh net on 30 Dec 2016 in the 5IP intake found large numbers of potential hosts or vectors of WSSV (Table 1), particularly in the south western (downwind) section of the main intake channel and the intake to pond 23 (Table 1). There was not enough time to properly enumerate the various types of crustaceans in samples E and F but all these samples remain fixed in 70% ethanol in the laboratory fridge at 5IP for later analysis.

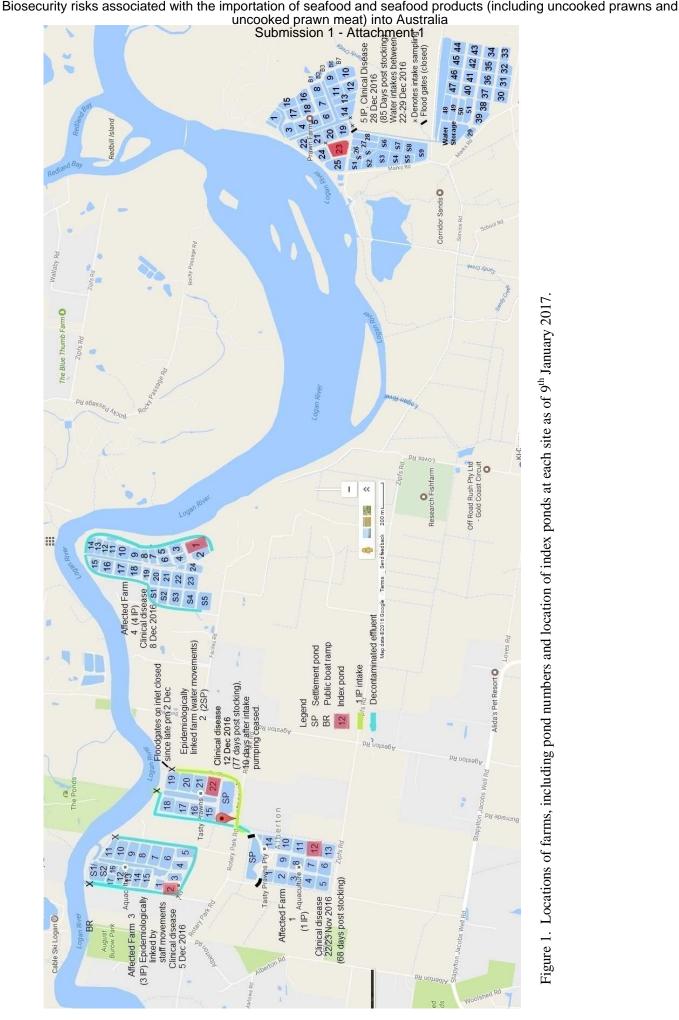


Figure 1. Locations of farms, including pond numbers and location of index ponds at each site as of 9th January 2017.



Figure 2. Location of WSSV positive mud crab (X) sampled on 23 December 2016 in the 7ARP discharge channel in relation to a nearby fishing area (red highlight). For details see Figures 6, 7. NOTE: for revised mudcrab location see Situation report #9.

Table 1. Plankton samples taken with a 15-20 µm mesh net on 30 Dec 2016 in the 5IP intake.

Sample	Time taken	Location	Result
A	4.48 pm	Southwest branch of intake near pond 19	N = 3 including 1 Lepostraca-like copepod 2 harpactacoid copepods
В	5.08 pm	Southern end of main intake channel	N = 22 including 11 Lepostraca-like copepods, 2 harpactacoid copepods, 2 Cladoceran-like, 2 cyclopoid copepod sp. 1, 4 cyclopoid (?) copepod sp. 2, 1 Zoea larvae
С	5.20 pm	Intake canal to ponds 23, 24	N = 164, including Harpactacoid copepods, cyclopoid copepod sp. 1 cyclopoid (?) copepod sp. 2, prawn nauplii, post larval prawns, Ostracod-like species, fish (mullet) larvae
D	7.00 pm	Southwest branch of intake near pond 19	N = 41, including 23 insect larvae, 10 harpactacoid copepods, 1 cyclopoid copepod sp. 1, 5 cyclopoid (?) copepod sp. 2, and 2 <i>Acetes</i> sp. (jelly prawn) larvae
Е	7.45 pm	Southern end of main intake channel	High diversity and numbers of plankton, including fish (mullet) larvae, <i>Acetes</i> (jelly prawn) larvae, Calanoid copepods, Mysids (?), zoea, etc. Too many to count in time available.
F	8.10 pm	Intake canal to ponds 23, 24	Species as per C, but too many to count in time available

- 4. After cleaning up the laboratory and storing the plankton samples in 70% ethanol in the fridge, I met with Biosecurity QLD field collection staff (Simon and Daniel) at 6.45 pm. The wind remained from the north east but was easing, and a slow moving storm front was approaching from the south, bringing a southerly change with it. While waiting for dark we discussed the original positive samples of 6 prawns from the river and Simon mentioned that they recalled such a sample and that it may have been taken from the 1IP inlet rather than from the river itself, but that he would recheck with Biosecurity QLD to ascertain whether this was the case.
- 5. After Simon and Daniel dropped off their crab pots they returned and launched a small boat to begin plankton tows in various locations within the 5IP intake canal. I walked to the southern end of the intake to retrieve my castnet and plankton nets left on site from the previous night and noted several king prawns still visible at site 3. With the wind still from the north (Figure 3) and now dark I walked back and conducted a cast net sample at site 6 at the very northern end of the intake (Figure 4). The results from 5 casts were 14 prawns, including 10 *P. plejebus*, 3 *Penaeus monodon*\* and 1 *Metapenaeus*, all of which were fixed in 95% ethanol then provided to Biosecurity QLD for analysis. A summary of the results from cast net sampling the 5IP intake with the wind from the North East is presented in Table 2 and Figure 4. The storm front then hit and wind direction changed to South East with gusts estimated 15-20 knots and pouring rain (Figure 3).

Table 2. Results of cast net sampling from sites 1-6 in the 5IP intake during northerly/north-easterly wind conditions. Results are sum totals from 5 random casts after at least one *P. plejebus* was visible on the surface.

Site	Time taken	Location	Penaeus plejebus	Penaeus monodon*	Metapenaeus	Total catch	% of total catch
1	7.30 pm 31/12/16	East side of pond 20	15	3	1	19	9.8%
2	8.00 pm 31/12/16	East side of pond 19	30	4	12	46	23.5%
3	8.30 pm 31/12/16	Southern end of main intake channel	41	4	7	52	26.5%
4	9.00 pm 31/12/16	North intersection of ponds 13, 14	8	0	5	13	6.6%
5	9.30 pm 31/12/16	Eastern end of east intake channel	39	7	6	52	26.5%
6	7.30 pm 2/1/17	Northern end of main intake channel	10	3	1	14	7.1%
Total		All sites	143	21	32	196	100 %

<sup>\*</sup> Note: identification subsequently revised to *Penaeus* spp., see Situation report #9.

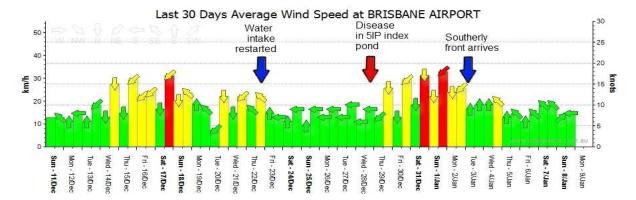


Figure 3. Wind records from past 30 days with events at 5IP superimposed.

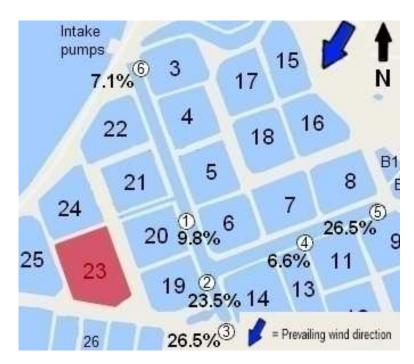


Figure 4. Results of cast net sampling from sites 1-6 in the 5IP intake during northerly wind conditions. The highest densities of prawns were at the downwind ends of the intake canals, especially in the south/westerly sampling sites (sites 2 and 3).



Figure 5. Results of cast net sampling from sites 1-6 in the 5IP intake during southerly wind conditions. The highest density of prawns was again at the downwind site, this time at the north end of the intake canal (site 6).

6. We drove back to the south end of the intake to sample site 3 and drop off the castnet and noted the king prawns were no longer visible as they left the edges of the canal and moved back into deeper water as the storm hit. In 5 blind casts a total of only 14 prawns were taken from site 3 (5 *P. plejebus*, 2 **Penaeus monodon** and 7 **Metapenaeus**) compared to 52 the previous night. All 14 were provided fresh to

Biosecurity QLD for analysis. Given the lightning and adverse weather conditions a unanimous decision was made to retire, and we stopped sampling, decontaminated boots and equipment, then left the farm at 10.15 pm.

# 3<sup>rd</sup> January 2017

- 7. On Tuesday 3<sup>rd</sup> January the morning was spent in communications and I participated in a Technical Advisory Group phone conference from 12.00-1.30 pm. Here it was confirmed by Mark Crane from AAHL that the mudcrab sample from the 7ARP outlet canal was the only positive sample from that location (Figure 5) and that subsequent samples from that location and from on farm at 7ARP were all negative, opening the way for 7ARP to start their harvest. Laboratory techniques for testing plankton and polychaetes and stress testing prawns collected from the intake at site 5IP were also discussed.
- 8. After the phone conference I departed for 5IP and arrived on farm at 4.45 pm where I spoke to site controller Ben who advised me that ponds 14, 42, 43 and 45 had been chlorinated (and pond 44 had been redosed). Weather was overcast, cooler (24-25°C) with 15-20 knots of southerly wind and occasional rain showers. I advised the site controllers that I would again be working with Biosecurity QLD field collection staff (Simon and Daniel) to take plankton and cast net samples from the 5IP intake, repeating previous work but this time with the southerly wind, to see if the distribution of crustaceans in the intake had changed with the different wind conditions.
- 9. While waiting for the sun to set I inspected the rocks within 30 meters of the 5IP intake at low tide and discovered several Sydney rock oysters ( $Saccostrea\ glomerata$ ) at the lower intertidal zone, which I sampled (n = 28), dissecting and fixing gills, palps and digestive gland from 17 (min shell length 34 mm, max 98 mm, mean 61 mm shell length, SD = 15.03) in 95% ethanol and placing them in the fridge in the 5IP laboratory, and providing the remaining 11 fresh and unopened to Biosecurity QLD for analysis.
- 10. Once the sun had set I brought in a new cast net and repeated the cast net sampling at the same 6 sites in the 5IP intake, as was done previously in the northerly wind conditions. The results from this are presented in Figure 5 and Table 3. While catch rates were down slightly from the previous days (possibly due to reduced prawn activity from the cooler weather), it was apparent that the distribution of prawns in the intake had shifted significantly, with the highest densities now being found at site 6, (which again was the downwind site in the southerly wind conditions) where 60 prawns were captured in 5 throws of the castnet (40.8% of all prawns captured) (Figure 5, Table 3). Prior to conducting the cast net sampling at each site I also collected plankton samples with the 15-20 µm mesh net and fixed these in 70% ethanol. Representative samples of the prawns captured (including samples of king prawns with melanised cuticular lesions on the abdominal segments from site 6) were also fixed in 95% ethanol.
- 11. Sampling was completed by 11 pm, after which all nets were left on site, boots were decontaminated, and the samples stored on site in the 5IP laboratory fridge. Upon leaving the laboratory at 11.20 pm I saw several live *P. monodon* on the ground near the cooker where they had apparently fallen out of a harvesting bin. I left the farm at 11.30 pm and returned home.

## 4<sup>th</sup> January 2017

12. On Wednesday 4<sup>th</sup> January part of the morning was spent with communications and the industry phone hookup. In the early afternoon I prepackaged, sealed and labeled prawn bait samples stored in the -20°C freezer at DigsFish and personally drove them to Eagle Farm where they were handed over for delivery for a testing program being co-ordinated by Matt Landos.

Table 3. Results of cast net sampling from sites 1-6 in the 5IP intake during southerly wind conditions. Results are sum totals from 5 random casts after at least one *P. plejebus* was visible on the surface.

Site	Time taken	Location	Penaeus	Penaeus	Metapenaeus	Total	% of total
	on 3/1/17		plejebus	monodon*		catch	catch
1	7.20 pm	East side of pond 20	11	4	2	17	11.6
2	7.54 pm	East side of pond 19	9	3	8	20	13.6
3	8.30 pm	Southern end of main	11	3	5	19	12.9
		intake channel					
4	9.13 pm	North intersection of	8	3	6	17	11.6
		ponds 13, 14					
5	9.45 pm	Eastern end of east	11	0	3	14	9.5
		intake channel					
6	10.20 pm	Northern end of main	40	6	14	60	40.8
		intake channel					
Total		All sites	90	19	38	147	100%

<sup>\* \*</sup>Note: identification subsequently revised to *Penaeus* spp., see Situation report #9.

### 5<sup>th</sup> January 2017

13. On Thursday 5<sup>th</sup> January the early afternoon was spent in an industry phone hookup. I was informed during the hookup that the intake canal at 1IP was chlorinated that day.

## 6<sup>th</sup> January 2017

- 14. On Friday 6<sup>th</sup> January in late AM we were informed by Barnaby Joyce that imports of green prawn products were suspended (for 6 months). I then visited the outlet canal next to the road down from Rocky Point Farm (7ARP) (Figure 2) where the WSSV positive mudcrab was sampled on 23 December. I arrived there at around 2.30 pm and interviewed Prawn Park worker Chris Woodberry who informed me that until the last 3-4 weeks (since signage and fences were erected), large numbers of people frequently fished a section of the outlet canal just upstream from where the positive mudcrab was sampled (fishing section, highlighted red in Figure 2). He advised me that the fishers generally used English as a second language and they would set up in groups along the canal most days of the week, line fishing using bait for bream and other regular estuarine target species, crabpots for targeting crabs and using cast nets for prawns. The section of the outlet canal where the mud crab was sampled also had signs of fishing activity (regularly used access tracks) and had been chlorinated (Figure 6), but the section used as a fishing area (Figure 7) still had many live fish (mainly mullet) and crustaceans (prawns) evident flicking in the shallows as well as small mud crabs and grapsid crabs which were visible along the canal edges. Both sections of the canal were narrow with minimal water movement, probably exchanging only on higher spring tides or when 7ARP ponds were drained, thus providing minimal dilution factor for any virus that may be present if imported prawns were used as bait. Indeed, it would seem prudent to fully sequence the WSSV from the mudcrab from the 7ARP outlet, to determine if its the same strain as the WSSV infecting the other farms. Given the large amount of fishing that historically occurred in the 7ARP drain, there may be a small chance the WSSV here was a separate introduction.
- 15. Just after 3 pm I arrived at 5IP, signed in at the decontamination checkpoint, decontaminated unused plankton nets that had been stored on the grass at the checkpoint using the chemicals provided under supervision of Biosecurity QLD staff, and moved the nets to the warehouse to dry. I then changed the fixatives, decontaminated, signed out and left 5IP, driving over to inspect the chlorination process in the 1IP intake.



Figure 6. Section of the 7ARP outlet canal on Friday 6<sup>th</sup> January, 14 days after a WSSV positive mud crab was sampled at this site. NOTE: for revised mudcrab location see Situation report #9.



Figure 7. Fishing section of the 7ARP outlet canal (red area from Figure 2) around 600 meters upstream of the section seen in Figure 6 where a WSSV positive mud crab was sampled. Well used access tracks and

discarded fishing gear suggested a significant amount of fishing activity had occurred at this site prior to erection of the biosecurity warning signs in late December 2016.

16. At the 1IP intake new advisory signs had been installed and Biosecurity QLD staff were in a boat checking chlorine levels (Figure 8). Large numbers of dead fish and crustaceans were visible, including greasyback prawns, jelly prawns, giant shrimp (Macrobrachium sp.), and small (8-10 cm CW) mud crabs, while fish included brown spot estuary cod (est. 2-3 kg), yellowfin bream, bony bream, silver biddy, mullet, glass perchlets, flathead, freshwater eels (est 5-6 kg), forktail catfish, Scatophagus sp., and butter bream. Examination of the bank next to the canal found large numbers of live graspid crabs in their burrows (Figure 9) and evidence of fishing in the form of access tracks near the intake (Figure 10). Luke Rossman then drove me to where the 1IP intake intersects the river at 5 pm. I then undertook a comparison between a new 15-20 µm plankton net and a new 100µm plankton net by running first the 100µm plankton net in the river, then the 15-20µm net through the same locations 15 minutes later. The net samples were fixed in 70% ethanol after which the nets were decontaminated in 300 mg/L chlorine for 1 hour. It was evident that much more phytoplankton and particulate matter was sampled using the 15-20 µm net (Figure 11). With a new castnet I then sampled the river at the intake from 5.30-6.30 pm to find mullet, numerous bream, and occasional butter bream and glass perchlets, as well as 15-20 small Metapenaeus sp., a dead mud crab and a dead prawn that probably had exited the floodgate from the chlorinated side. All of the crustaceans collected were fixed in 95% ethanol. The castnet was left at the rear gate to 2IP, after which we drove to the 2IP decontamination point, decontaminated boots, equipment and the car at the checkpoint and stored the plankton and crustacean samples on site in the 1IP fridge. I then left the farm at 7 pm and returned home.

## 9<sup>th</sup> January 2017

- 17. On Monday 9<sup>th</sup> January I phoned Simon who was collecting for Biosecurity QLD and he informed me that several more ponds were now confirmed as WSSV positive on 5IP, including ponds 1, 10, 15, 16 and possibly others (to be confirmed).
- 18. I was asked to meet with Darryl McPhee and Ron Boswell from the FRDC and caught up with them at Jacobs Well at 12.30 pm. After a briefing we proceeded to the 7ARP outlet canal by 2 pm where they observed the sites where the fishing and WSSV positive mud crab had occurred (Figures 6, 7). At the fishing site (27°43.480 S, 153°20.511 E) Biosecurity QLD had a crab pot in the water, but around 5 meters away a juvenile mud crab was observed moving in a conspicuous fashion along the canal edge (Figure 12), suggesting that dip netting may be a more effective method of sampling crabs showing unusual behaviour.
- 19. After inspecting the 7ARP outlet we visited 5IP, signed in at the checkpoint and had a meeting with Noel, Alistair, and Nick in their office from 2.30 till 3.30 pm. After signing out we then visited the office of 7ARP to have discussions with Serena and Murray Zipf from 3.30-4 pm. After that meeting we then inspected the 1IP intake from the road where we were met for discussions by Dan, Simon and Ian Rossman. Biosecurity QLD staff had cleaned up most of the dead fish from the intake, however live graspid crabs were still evident on the canal edges (Figures 9,10). After finishing the meeting at 5.30 pm, I returned home to begin writing sitrep#8.

### 10<sup>th</sup> January 2017

20. On the morning of Tuesday  $10^{th}$  January I contacted the CVO and suggested the need to conduct plankton tows in the 7ARP canal as well as in a WSSV infected pond at 5IP in order to compare effectiveness of 15-20 vs  $100~\mu m$  (DigsFish) vs  $250/500~\mu m$  (Biosecurity QLD) plankton nets for detecting WSSV carriers. I also indicated the potential for placing oysters in the 7ARP canal, 5IP intake and in an infected pond at 5IP to determine their suitability as sentinels for detecting WSSV in the water. Such an experiment would help confirm if local species of shellfish can indeed concentrate WSSV, as has been recorded overseas.



Figure 8. The 1IP intake was chlorinated on 5th January 2017 and re-checked on the 6<sup>th</sup> January. Signs had recently been erected informing road users the area was a biosecurity control area.



Figure 9. Examination of the bank next to the 1IP intake canal found large numbers of live graspid crabs remain in their burrows after chlorination of the water.

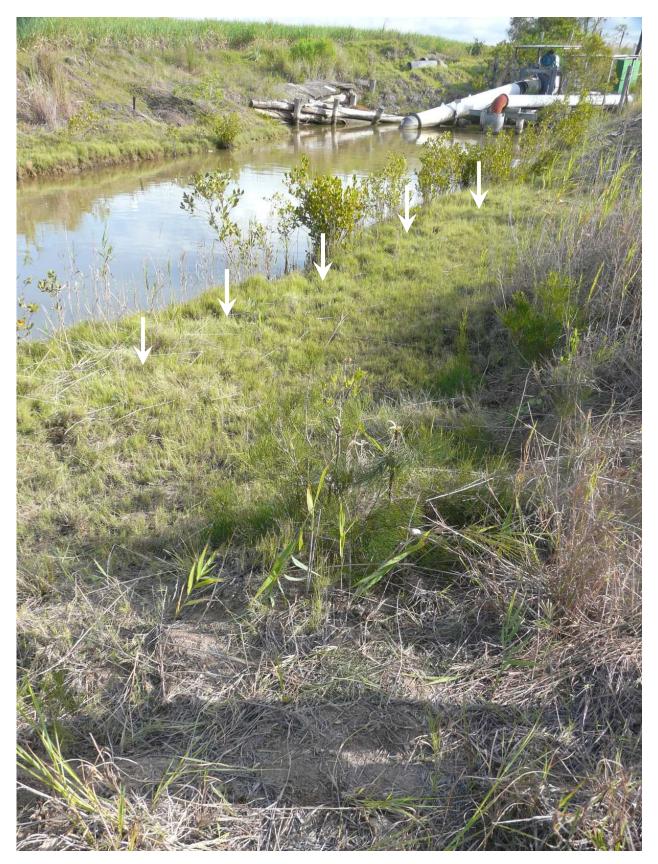


Figure 10. Evidence of fishing activity in the 1IP intake canal in the form of access tracks towards a bank immediately adjacent to the water intake pipes. Location of grapsid crab burrows arrowed.



Figure 11. Comparison between samples collected from the junction between the 1IP inlet and the Logan River in a 15-20  $\mu$ m plankton net (left) and a 100 $\mu$ m plankton net (right). Much more phytoplankton and particulate matter was sampled using the 15-20 $\mu$ m net, suggesting it may be a useful method of sampling environmental/particle associated WSSV.



Figure 12. A juvenile mud crab observed moving in a conspicuous fashion along the edge of the 7ARP outlet canal edge on 9<sup>th</sup> January 2017. Failure to collect this crab in the nearby crab pot situated 5 meters away suggests that dip netting may be a more effective method of sampling crabs showing unusual behaviour.

# Appendix 9. WSD outbreak situation report #9. 13 Jan 2017

This is a summary of developments along the Logan River since my last report of 9<sup>th</sup> January 2017. The current situation with respect to infected premises and significant sampling events can be found in Figure 1 (over page).

## 12th January 2017

1. On Thursday  $12^{th}$  January 2017 I attended the Australian Prawn Farmers Association Executive Meeting on the 5th floor of the DPI Building. The meeting was an opportunity for Biosecurity QLD to update the industry on the status of the response to the WSD incursion on the Logan River. Biosecurity QLD reiterated their aim to try to return to freedom from White Spot Disease, then provided updated information on the various findings from their activities and investigations. Some of the information on the CT values of samples taken from *Penaeus monodon* sampled from WSD affected farms is contained in Table 1. Some of the information presented on the CT values of crustaceans sampled from the wild (n = >6900) are contained in Table 2.

Table 1. CT Values for WSSV positive results from *P. monodon* sampled from infected farms on the Logan River.

Farm	Date positive confirmed (BSL)	AAHL confirmed	CT value (range)
1IP	30 Nov 2016	1 Dec 2016	14.00-21.13
2IP	13 Dec 2016	15 Dec 2016	17.1-31.5
3IP	6 Dec 2016	7 Dec 2016	Not reported
4IP	9 Dec 2016	9 Dec 2016	20.2-43.04
5IP	29 Dec 2016	29 Dec 2016	14.00- 26.00

Table 2. CT Values for WSSV positive results from wild prawns and crabs sampled from the Logan River and its drains.

Location	Sampling location details	Date Sampled	Species	CT value
2 IP (adjacent)	Trawl #2	5 Dec 2016	4 Metapenaeus	37.5-39
2 IP (downstream)	Trawl #3	5 Dec 2016	1 Metapenaeus,	37.5-39
			1 glass shrimp	
7ARP	Drain pond next to road	23 Dec 2016	Scylla serrata	40.46

2. The location of sampling of the 6 wild prawns that were WSSV taken from the Logan River was confirmed to be in the Logan River itself in beam trawl shots #2 and 3 which were taken adjacent to the inlet of 2IP on 5 December 2016 (Figures 1, 2). The vast majority of the locally sourced bait samples tested to date (n = >2000) were negative, only one early batch from the Gem Bait and Tackle being indeterminate (WSSV positive on some tests but not others). The location of capture of the WSSV positive mudcrab in the outlet channel from 7ARP was confirmed to be at the south-western end of the channel (Figure 3), not the eastern end as I was lead to believe (and therefore incorrectly reported in my previous SitRep Report (#8) -my apologies). Serena Zipf did, however, confirm that a large amount of recreational fishing effort also occurred at the site where the mudcrab was sampled as it was immediately adjacent to the road (Figure 3). Areas of recreational fishing access where fishing activity was observed to regularly occur either by myself or through statements from interviews with local residents are shown as red shaded areas in Figures 2 and 3.

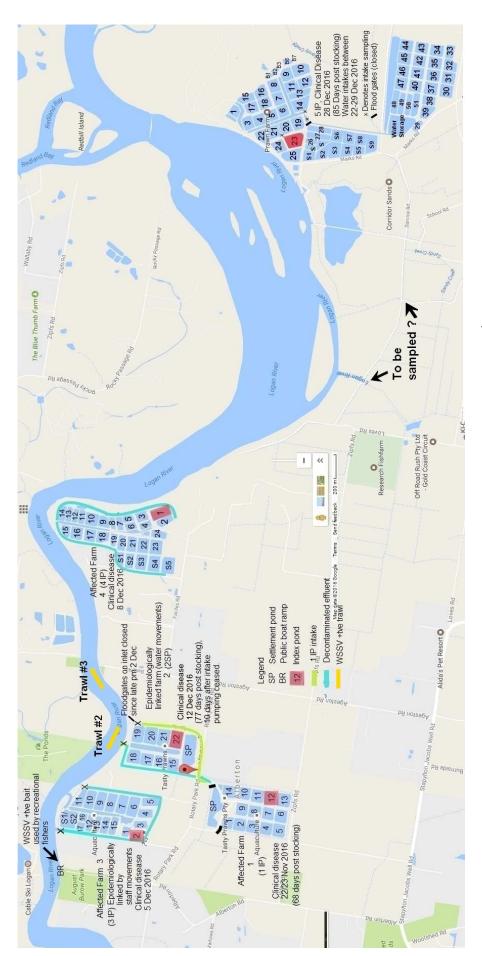


Figure 1. Locations of farms, including pond numbers and location of index ponds at each site as of 13th January 2017.



Figure 2. The location of trawl shot #2 taken in the Logan River adjacent to the inlet of 2IP on 5 December 2016 in which 4 wild *Metapenaeus* were detected. A third shot around 500 M downstream (east) of 2IP also collected a single positive *Metapenaeus* and a single positive glass prawn (*Acetes* sp.). Red shading shows public access points on the 1IP inlet where recreational fishing has been observed.



Figure 3. Corrected location of WSSV positive mud crab (X) sampled on 23 December 2016 in the 7ARP discharge channel in relation to a nearby fishing areas (red highlight). For details see Figure 4.



Figure 4. Direct access off public road to 7ARP outlet /intake where WSSV positive mud crab was sampled from the outlet pond (arrow) (= X in Figure 3).

# 13<sup>th</sup> January 2017

3. On Friday 13<sup>th</sup> January 2017 I attended a meeting at the Yatala Motel where prawn farmers met with international experts to discuss their options moving forward for the 2017/18 season. One significant development from this meeting was a report from Warren Truloff that fisheries officers whom he had met near his property had informed him that in the month or so since the area closure had been implemented for recreational fishers on the Logan River, the fisheries officers had detected 6 groups of fishers near the Alberton Boat ramp (top left of Figure 1) using imported raw prawns as bait. Warren reported the officers also indicated that of the 6 bait samples confiscated and tested, 2 had returned "strong positive" results for WSSV infection. While these figures remain to be verified, the fact that recreational fishers operating near the affected prawn farms were caught using WSSV infected imported prawns for bait was officially confirmed by the Federal Department of Agriculture and Water Resources in a media statement as follows;

"In the course of our investigations, the department did come across recreational fishers using imported prawns labelled for human consumption for bait in the Logan River. Subsequent testing of the product did return positive results for the virus. What this tells us is that fishers using infected imported prawns for bait is one possible pathway for this disease to get into our river system and onto prawn farms and is why prawns imported for human consumption should never be used for bait." The Federal Department also stated "We are still looking at a number of pathways that may have resulted in the white spot disease incursion in Queensland, including imported feed or probiotics, contaminated equipment, or even discarded uncooked prawns - or bits of prawns - that were purchased to eat".

- 4. Nick Moore also mentioned at the meeting that he expects the last of the ponds on his farm to be chlorinated today. After the meeting I was guided by Luke Rossman to two creeks/drains on their cane property between 4IP and 5IP that appear to be strategically positioned as sampling locations where WSSV may have possibly been concentrated in recent times during northerly wind events. The locations were a floodgate /bridge over sandy creek (27°42.978 S, 153°18.176 E) and a drain named Flood Structure 34 (27°43.153 S, 153°18.585 E). The Biosecurity Sampling team were contacted to see if they had sampled these locations. They had not, but informed me they expect to be able to sample them over the next few days.
- 5. After viewing these potential sampling sites, I drove to the 7ARP outlet where the exact location where the WSSV infected mudcrab was ascertained with Murray Zipf it was collected from the outlet pond nearest the public road access point (Figures 3, 4) which was a body of water around 80 x 20 meters in dimension with minimal/no water flow. Again, both Murray and Serena informed me that members of the public regularly fished both the inlet and outlet drains, including since the warning signs were erected. As the outlet was chlorinated I took 2 plankton samples (1 x 100  $\mu$ m and 1 x 15-20 $\mu$ m) from the southern end of the inlet drain, fixed the samples in 70% ethanol and left these in the fridge at the office at 7ARP, before returning home.

#### **Erratum**:

6. It as been bought to my attention that there is a possibility that the tiger prawns sampled from the 5IP intake during earlier crustacean distribution studies (reported in SitRep Reports #7 and #8) were possibly not *P. monodon*, but could be *P. esculentus* or *P. semisulcatus*. Until such time as more detailed taxonomic study of the ethanol fixed samples archived at 5IP are undertaken, the prawns referred to as *P. monodon* in Table 2 and Figure 8 in SitRep Report #7, and Table 2 in SitRep Report #8 should be referred to as *Penaeus* sp. until their identities are confirmed.

# Appendix 10. Ridley position statement on prawn feed 5<sup>th</sup> Dec 2016



December 5th, 2016.

#### RIDLEY POSITION STATEMENT

### "WSSV (White Spot Syndrome Virus) Incident in the Logan Prawn Farming Region"

WSSV is a highly infectious disease of crustaceans, including farmed prawns. In farming practice, it is spread via the transfer of infectious animals through broodstock, nauplii, post larvae or other life stages of the prawns. There are many other modes of disease transfer including the movement of water from infected ponds, movement of dead or dying animals via other wildlife (e.g. birds), use of infected animals as bait and the movement of people and farm equipment between farms.

There is no evidence of transfer of WSSV via feed. WSSV is highly heat sensitive, which means that it is deactivated by heat1,2 very rapidly. During production of feed, cooking temperatures of Ridley prawn feed is between 85-95°C for between 30-45 minutes. This is more than sufficient to deactivate any virus present. In addition, Ridley policy is not to use any farmed crustacean products (e.g. shrimp head meal) in their formulations in order to mitigate any risk, however small this risk may be. All feed is packed in clean new bags.

Ridley's current freight arrangements heading to the North of Qld, is via rail in the first instance, and as such there is a clear separation of feed delivery from the Logan area to other prawn farming regions. In the few instances where farms arrange their own feed deliveries, these farms have been asked to notify their carriers to not use their vehicles in the Logan farms area as well as heading North to other farms...

For further information, please contact Dr Richard Smullen or Dr Matt Briggs

- Morris et al. 2007 Peer Review of the Biosecurity Australia's Revised Draft Generic Import Risk Analysis Report for Prawns http://www.agriculture.gov.au/SiteCollectionDocuments/ba/animal/prawnsubmissions/SIAA Peer Review.pdf
- 2. Mathias Corteel, 2013. White spot syndrome virus infection in P. vannamei and M. rosenbergii: experimental studies on susceptibility to infection and disease. Dissertation submitted in fulfillment of the requirements for the degree of Doctor of Philosophy (PhD) in Veterinary Sciences

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# Appendix 11. Minutes of DAWR-Industry WSD Phone Conference

WHITE SPOT DISEASE	Meeting No	01	
QUEENSLAND 2016	Location	TC	
INCIDENT MANAGEMENT TEAM INDUSTRY MEETING SUMMARY	Date	15 2016	December
INDESTRI MEZIMO SOMMARI	Time	2.30pm	n – 4pm

Attendees:

Chaired by: Ian Thompson

Name	Division / Branch or Jurisdiction
Ian Thompson	Department of Agriculture and Water Resources
Robyn Martin	Department of Agriculture and Water Resources
Tim Chapman	Department of Agriculture and Water Resources
Gordon Neil	Department of Agriculture and Water Resources
Terri McGrath	Department of Agriculture and Water Resources
Brett Herbert	Department of Agriculture and Water Resources
Giulia Porro	Department of Agriculture and Water Resources
Helen Jenkins	Australian Prawn Farmers Association
Annie Jarrett	Northern Prawn Fishery Industry Association
Eric Perez	Queensland Seafood Industry Association
Aaron Irving	National Aquaculture Council
Trisha Beattie	New South Wales Seafood Industry Association
Felicity	Shark Bay Prawn Fishery
Grahame Turk	Sydney Fish Markets
Johnathon Davey	Seafood Industry Victoria / National Seafood Industry Association
Alex Ogg	Western Australia Fishing Industry Council
Matt Landos	Future Fisheries Veterinary Services
Ben Diggles	DigsFish
Jim Thompson	Queensland Department of Agriculture and Fisheries
Scott Spencer	Queensland Department of Agriculture and Fisheries
Patrick Hone	Fisheries Research and Development Corporation
Peter Horvat	Fisheries Research and Development Corporation

#### **Discussion**

The Department of Agriculture and Water Resources (the department) provided attendees with an update on the incident management process as well as information about the white spot disease (WSD) and the international and national experience.

Queensland Department of Agriculture and Fisheries (QDAF) provided attendees with a summary of the current situation and recent developments. QDAF have established a state coordination centre as well as a local control centre. Around 70 staff from the Queensland government are involved in the response. Approximately 15 people have been deployed to Queensland from other jurisdictions to assist. The current response is focusing on eradicating WSD and disposal and decontamination of the infected premises. Movement restrictions are in place along the Logan and Albert Rivers.

Attendees discussed tracing the source of the infection. It appears that the predominant message circulating amongst industry as well as in the media is that the WSD was transmitted through raw frozen prawns imported for human consumption. Tim Chapman advised that the source of infection was still unknown and all possible pathways were being considered and investigated. The assumption that the infection could have come from imported prawns was premature and at this stage the source of the white spot disease outbreak is not known.

Tim Chapman provided attendees with information about prawn consignments that have arrived in Australia. Between May and December 2016, 448 consignments have arrived in Australia, of these -73 consignments had tested positive for WSD and were re-exported. Tim Chapman clarified that as part of the prawn import conditions, 100 per cent of consignments are tested for WSD when arriving into Australia.

Attendees agreed that maintaining positive communication and media was essential throughout the WSD response. QDAF has been engaging directly with industry associations (both for farmed prawns and wild caught) and with the impacted premises owners. QDAF advised that the Queensland Seafood Industry Association is part of the Queensland incident response body. QDAF and the department agreed to continue engaging directly with the Australian Council of Prawn Farmers, the Australian Prawn Farmers Association, the Queensland Seafood Industry Association, the National Seafood Industry Association and Sydney Fish Markets. QDAF has also been engaging with recreational fishers through Sunfish.

Industry associations expressed concerns about scaring the public away from prawns and seafood generally, particularly at this time of year. Attendees agreed that it would be best to keep the communication about the WSD and movement controls to the local area. Industry associations agreed that working together and focusing on positive messaging would be beneficial.

Industry associations also raised concerns about the sale of bait and the processes in place to trace potential infections. QDAF advised attendees that were sampling bait from the local suppliers. Attendees discussed the issue of regulating the bait industry.

QDAF advised that the issue of industry assistance is being considered.

The overall feedback from industry was that the teleconference was useful.