

**Submission to the inquiry into the biosecurity risks associated with the importation of seafood and seafood products (including uncooked prawns and uncooked prawn meat) into Australia**

**Submitter contact details**

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Dear Sir/Madam,

Re: Submission to the inquiry into the biosecurity risks associated with the importation of seafood and seafood products (including uncooked prawns and uncooked prawn meat) into Australia, I would be grateful if you could consider the following submission.

**Summary:** This submission provides **clear scientific evidence based on DNA sequences that the source of the White Spot Syndrome Virus infection in Logan River prawn farms was from overseas** and not from long term endemic strain/s. This submission also reports that hypervariable DNA regions ("genetic fingerprints") of WSSV have been identified at USC and sequencing of these DNA regions, currently underway, will likely identify precise overseas regions whose WSSV strains are most closely related to that associated with the Logan River outbreak.

**Terms of reference addressed:**

*TOR i. Any related matters.*

I assume an understanding of the **source of the infection**, whether local or overseas, and if from overseas, from which country or region would come under *TOR i "any related matters"*.

*Outcome if this matter is resolved:* It is likely that knowledge of the source of the infection (presently unknown) would inform the industry how to best prepare for the future, for example, if the infection is from an endemic local source then the industry may need to develop permanent expensive on farm facilities to stop the entry of the White Spot Syndrome Virus (WSSV) into the farms, whereas if the source is determined to be from overseas, then perhaps investment to strengthen Australia's border defences would be more cost effective to prevent future infections.

**My background and relevance**

I am an Associate Professor at the University of the Sunshine Coast and my research focusses on genetics and aquaculture. I lead a group of 12 staff and PhD students and part of our research concerns with viruses in prawns and the use of DNA sequence technologies to resolve matters such as the origin of infections and matters of taxonomy of closely related species. I and my group have the experience, academic capability, technical skills and international standing, demonstrated by our many "peer reviewed" international publications, to resolve the source of white spot infection in Australia, public good information that could help the industry make informed decisions how to rebuild their industry in a cost-effective manner. Note: as an employee of USC, I am at liberty to make public comment within my field of expertise (such as the source of WSSV) but my comments do not represent any "USC" corporate position.

**Australia WSSV DNA sequences are identical to overseas DNA sequences, ruling out the possibility the Logan River infection was due to a long term endemic form of WSSV.**

So far we have obtained WSSV DNA sequences from nine different Australian prawn individuals; samples were from a Logan River aquaculture farm and taken at the time of the WSSV outbreak. Samples had white spots on the carapace (shell) which is characteristic of WSSV infection. These are

the first Australian WSSV DNA sequences ever reported and released to the public. The sequences from the Logan farm are all 100% identical to each other. *For most of the different regions of the genome tested*, the Australian sequences have a 100% match to some overseas samples, and a very high homology with all overseas samples. If the Australian WSSV in the Logan River originated from an endemic species (something that had been present in Australia for many “generations”) we would expect to see DNA nucleotide mismatches between the Australian DNA sequences and the overseas sequences throughout the genome because DNA sequences change/mutate over time, and separated populations diverge. Such mismatches were not observed for most DNA sequences and *most regions* of the genome; any mismatches in most genome regions were very minor. In scientific language, we would reject the hypothesis that the Logan River WSSV outbreak came from a long term endemic resident, rather, by far the best hypothesis that fits the data is that the Logan River WSSV is a very recent arrival from overseas.

#### **Are we able to find the precise overseas origin/region source of infection?**

*Justification and outcome:* While we have resolved by normal standards of scientific enquiry that the source was from overseas, there may be value in determining a more precise regional source of the infection, knowledge that may help us to understand how the WSSV entered into Australia; this new knowledge may assist in the prevention of future infections.

*Preparation:* To resolve this matter, first we collaborated with Dr Alexie Papanicolaou of the Hawkesbury Institute for the Environment, Western Sydney University to find very special regions in the WSSV genome that were hypervariable (i.e. regions of high mutability that make excellent “DNA fingerprints”). Our testing and DNA sequencing has shown we now have found a number of hypervariable and highly mutable DNA regions, and thus we now have the tools to identify the precise source of the infection.

*Sampling at USC:* We collected DNA samples from overseas animals (including those from current overseas WSSV outbreaks) and from imported processed prawns (frozen spring rolls etc.) currently available (as of May 2017) in Australian shops (the retail items usually state country of origin but we cannot guarantee the provenance of the retail samples). In addition to the recently collected DNA samples, we hold in our archives thousands of prawn samples from Australia and around the world from many countries which are available for testing.

*Results:* High quality DNA sequences from conserved and hypervariable “fingerprint” regions were obtained from overseas DNA samples and the local Australian retail samples. For some overseas countries, we did not yet find any matches with the Logan River samples (neither with our own overseas samples nor with DNA sequences published by other scientists for those countries). Similarly, some retail DNA sequences did not match the Logan River samples. Conversely, we have found nearly exact matches for all six hypervariable regions of the genome so far assessed with just one overseas region. Notwithstanding further testing of overseas samples which will be ongoing, we believe we now have a match (analogous to matching bullets from the same gun), and our research will shift focus to discovery of the exact pathway of entry (we need to find the “gun”); accordingly our research will now focus on testing Australian retail samples from the “region of interest”.

#### **Where can other scientists view our DNA sequence data?**

All our DNA sequences are being lodged with an international DNA sequence database called “GenBank” and as they are processed by GenBank, they will become available to the public. Some GenBank references numbers are MF287678, MF161441, MF161442, MF161443, MF1614410, MF161440 and MF348242. Scientific papers are also in preparation to be sent for international peer review and publication.

**Conflict of interest declaration**

This research was not supported financially by any entity outside of USC. The operating and technician costs were funded solely by the Aquaculture Genetics Group at USC, using facilities provided by USC. The above written submission was prepared entirely by Associate Professor Wayne Knibb, but based on work carried out the Aquaculture Genetics Group at USC.

**Acknowledgments**

We thank Logan River prawn farmers for kindly providing samples for testing.

Thank you for the opportunity to express my views,

Kind regards  
Wayne Knibb  
Assoc. Prof. USC  
Aquaculture and Genetics