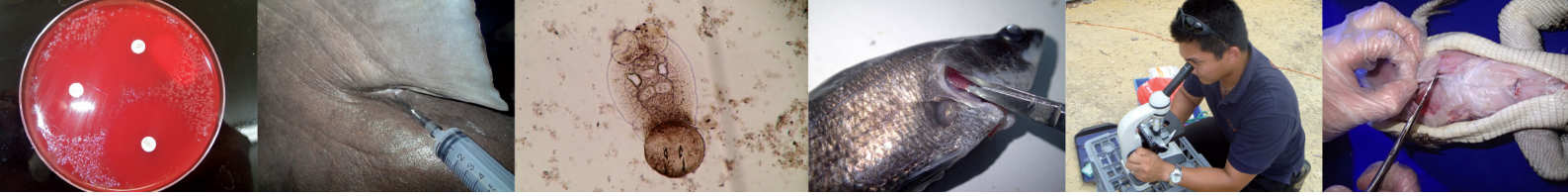


Proof Committee Hansard – SENATE

The Fish Vet

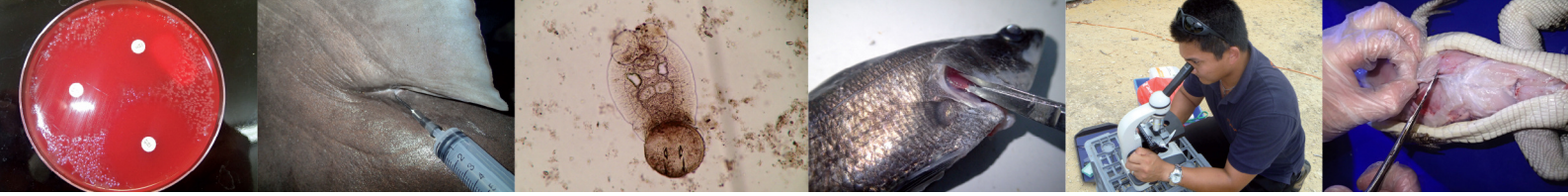
Only two areas for editing, included in attachment:

Page 8

Dr Loh: Cattle are highly susceptible to botulism. The dead fish will initially produce ~~it~~type E toxin. ~~It~~ This only affects fish and birds, and possibly~~. It wil~~l also affect humans. And~~, but,~~ if birds eat the rotting fish and they subsequently die, they will likely produce ~~botulism~~ botulinum type ~~B~~ C toxins. The type ~~E~~ C toxin can kill~~s~~ cattle, sheep, horses and a whole heap of different birds.

Page 11

Dr Loh: I don't believe this has been published in a peer-reviewed journal. Also there were a couple of instances where, say, silver perch were injected with the virus and they found the virus by PCR in the gills. They gave the Murray cod a bath and then they found the virus in the kidneys. This tells me that the fish has actually taken on the virus and circulated it systemically. In terms of the mortality, it's very high. With the normal carp that's captured ~~in~~ from the wild, in a bath experiment the mortality rate was between 65 per cent to 80 per cent; silver perch was 35 per cent; Murray cod was 24 per cent; golden perch was 42 per cent; galaxias was 82 per cent; and rainbow trout was 43 per cent. In the injected ones, the carp mortality rate was between 40 per cent to 75 per cent; silver perch was ~~65~~ 55 per cent; Murray cod was about 35 per cent; golden perch was 37 per cent; galaxias were too small for injection; and the rainbow trout mortality rate was 100 per cent.



Supplementary Submission

Further information on two issues as requested by Senators:

1. Botulinum toxin
2. Mortality rates in experimental transmission trials by the NCCP

1. Botulinum toxin

Cattle are very susceptible to the botulism toxin. Toxin production occurs in an anaerobic environment, with moisture and an optimum temperature of around 23 °C (15-35 °C). All these conditions can be found in a rotting carcass.

Virbac's Singvac 3 Year Single Shot Bivalent Botulinum Vaccine for Cattle product information states, "It is caused by the ingestion of Cl. botulinum toxin in feed (especially mouldy hay, decaying vegetation and the carcasses of dead cattle, sheep and rabbits) or from contaminated water."

The toxin itself may be stable for several days in untreated water, especially in acidic conditions. In a carcass, the toxin can last for a year at 30 °C.
Infection has also been known to occur by drinking water or consuming feed that has been contaminated by animal carcasses and rotting feed (mouldy or decomposing hay or silage).

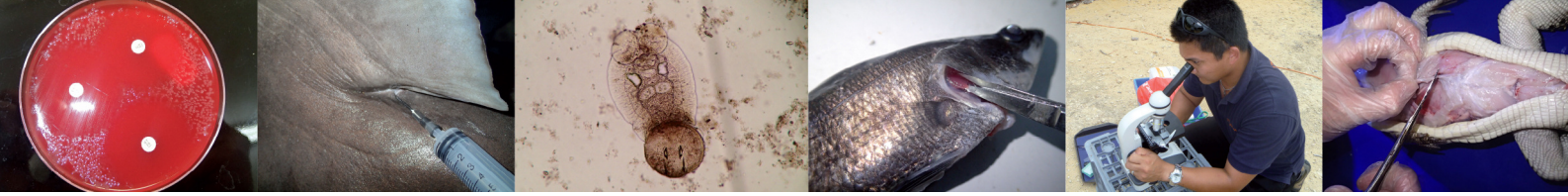
Regarding the spores, they are highly resistant in sediments, persisting for decades until conditions (anaerobic habitats as occurs in and around carcases) are right for proliferation.

<source: https://www.nt.gov.au/__data/assets/pdf_file/0017/233252/651.pdf>

2. Mortality rates in experimental transmission trials by the NCCP

This publication determined the course for NCCP's direction - McColl and Crane (2013) ***Cyprinid herpesvirus 3, CyHV-3: its potential as a biological control agent for carp in Australia.*** Invasive Animals Ltd <source: https://www.pestsmart.org.au/cyhv3-potential-as-a-biological-control-agent-for-carp-in-australia/>.

I found the report difficult to follow because the style of reporting differed between experiments. It was difficult to know how many fish were used in every experiment, and there was no detail as to the water quality parameters that were monitored, and lack details of experimental design. There were many unexplained deaths that were not investigated sufficiently. I would have thought this would have been a criteria set by the Animal Ethics Committee. There were numerous other irregularities in the work, but here we detail issues with high mortality rates in non-target species, and inconsistent approach to interpreting data.



From what I could decipher, results for mortality rates following exposure to CyHV3 by bath and injection, are tabulated below.

	Carp	**Silver Perch**	**Murray Cod**	**Golden Perch**	**Galaxias maculatus**	**Rainbow trout**
Bath	64, 79%	35%	24%	42%	82%	43%
Injected	40, 67, 75%	55%	35, 100%	37%	*Not trialled*	100%

The NCCP relied heavily on molecular testing to determine whether fish died from CyHV3 because in the carp examined, "Histologically, there was a paucity of lesions." Lesions were only seen in a single carp.

Mr Falconer mentioned the NCCP created their own definition for what was deemed positive and negative for their transmission studies. A lower Ct value means there is more DNA present; so as a general rule used by most laboratories, Ct of 36 or less is positive, Ct of between 37-45 is considered indeterminate (and required re-test), and Ct of 46 or greater is negative. The NCCP stated that for their experiments, Ct of 37 or greater is considered negative. The NCCP's report did the NCCP arrive at this?

Through the publication, no Ct values were mentioned for those that were deemed "negative". In an ordinary laboratory, results that fall between Ct of 37-45 would require re-testing, to ensure there were no inhibitors present that could produce an artefactually low Ct value. Consequently, the NCCP may have oversimplified their definition of positive versus negative, and this gave bias to the interpretation of results.

But then the NCCP disregarded their definition of "positive" when 22% of the dead/moribund injected silver perch, were positive for viral DNA in the gills with average Ct of 34.8-36.4, and one healthy silver perch was positive in the gills with Ct of 34.8. And then in the bath experiment with Murray cod, 24% of fish died, and viral DNA was found in 2/15, dead/moribund cod's kidneys with average Ct value of 35.4-35.6. To me these results are suggestive of viraemia and possibly a carrier status (this would have trade implications). Instead, the NCCP disregarded all these positives as being a contaminant.

With such high variability in mortality rates as shown by NCCP's experiments with Australian carp, it suggests to me that CyHV3 may not deliver the intended kill rates. The level of mortalities and positive results in non-target species does not give me confidence that CyHV3 is as safe an option as promised. The possibility of causing deaths in non-target species, or even a carrier status will have great ramifications.

Yours sincerely,

Dr Richmond Loh

DipProjMgt, BSc, BVMS, MPhil (Pathology) Murdoch, MANZCVS (Aquatics & Pathobiology), CertAqV.

Aquatic Veterinarian & Veterinary Pathologist