

Ranavirus in Australian Reptiles



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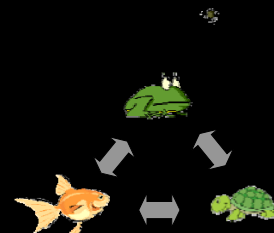
Ranaviruses

Implicated in the world-wide decline of amphibians and mass mortalities in both aquaculture and wild fish stock.

Reported to infect hosts across classes.

This ability has the potential to compromise prevention and control measures, since amphibians, reptiles and fish may act as reservoir species for each other.

Different hosts - similar virus



Reptile hosts for ranaviruses

Box turtle	USA
Soft-shelled turtle	China
Central Asian tortoise	USA
Gopher tortoise	USA
Hermann's tortoise	Switzerland, UK
Four-horned chameleon	UK
Gecko	Germany
Green tree python	Australia (ex-Irian Jaya)
Burmese star tortoises	USA



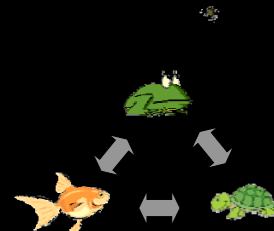
Bohle iridovirus - BIV

Isolated from newly metamorphosed frogs that died (*Limnodynates ornatus*).

Caused 100% mortality in barramundi (*Lates calcarifer*) – a native fish

Any effect on native reptiles?

Different hosts - similar virus



Bohle iridovirus in Australian reptiles

ELISA to test for past exposure

Serum survey

Experimental infection

Criteria:

Animals from a freshwater environment
in contact with fish & frogs

Not too dangerous...
poisonous or otherwise

Green tree snake



Brown tree snake



Keelback snake



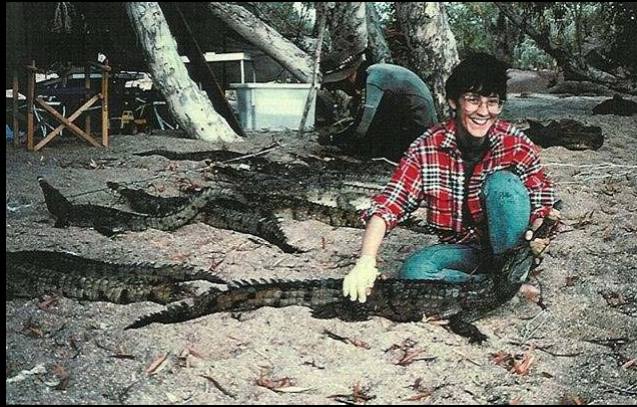
Kreffft's river tortoise



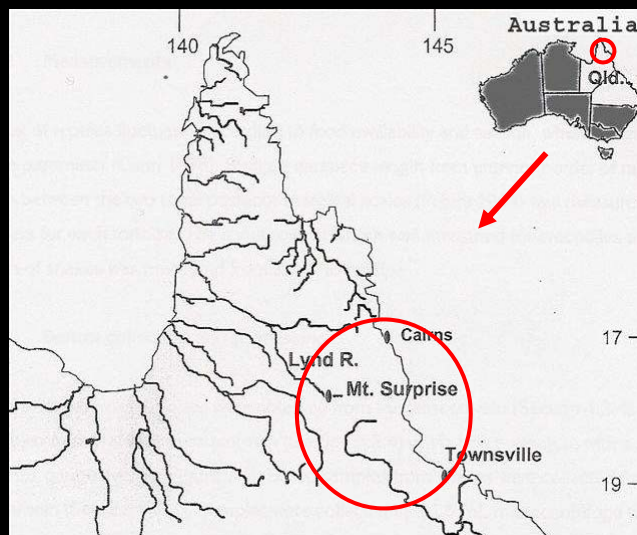
Sawshelled tortoise



Freshwater crocodile



Where in the world... Study area



Animals in the project



Blood sampling



Tail vein



Snake in a bag



Turtle femoral vein

Antibody ELISA

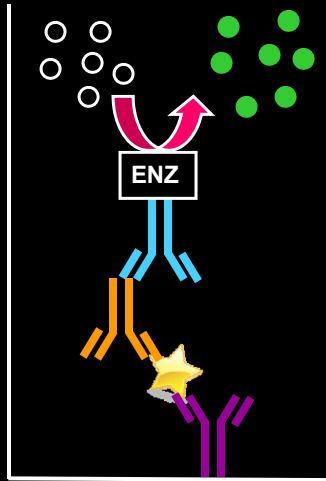
Chromogen and substrate

Enzyme-conjugated goat α Rabbit Ab

Rabbit α BIV PAb

Reptile α BIV PAb

TEST SERUM pos

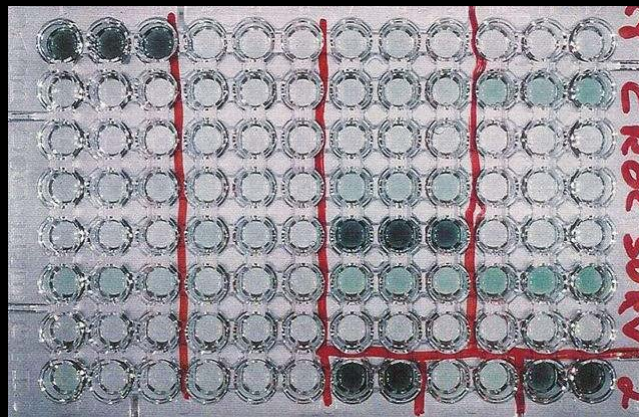


Coloured product

BIV antigen

Antigen capture ELISA with reptilian test sera

Serology survey

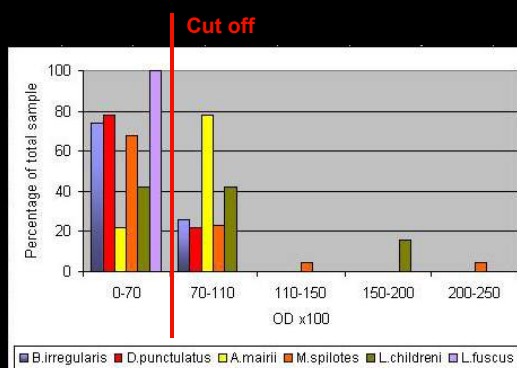


Ranavirus serology in reptiles

	<i>n</i>	%+ve
• <i>Elseya latisternum</i>	86	61
• <i>Emydura krefftii</i>	52	36
• <i>Crocodylus johnstoni</i>	63	25

Snakes

• <i>Boiga irregularis</i>	19
• <i>D. punctulatus</i>	19
• <i>Amphiesma mairii</i>	9
• <i>Liasis childreni</i>	7
• <i>Morelia spilotes</i>	21
• <i>Liasis fuscus</i>	2



Challenge trials

Animals were inoculated IC
and observed over a 30 day period

- *Crocodylus johnstoni* (16) none died
- *Boiga irregularis* (10) none died
1 +ve virus 28d
- *Dendrelaphis punctulatus* (6) none died
- *Amphiesma mairii* (7) none died
- No controls died or were positive for virus isolation

Challenge trials

Adults

- *Emydura krefftii* (12) none died
- *Elseya latisternum* (12) none died

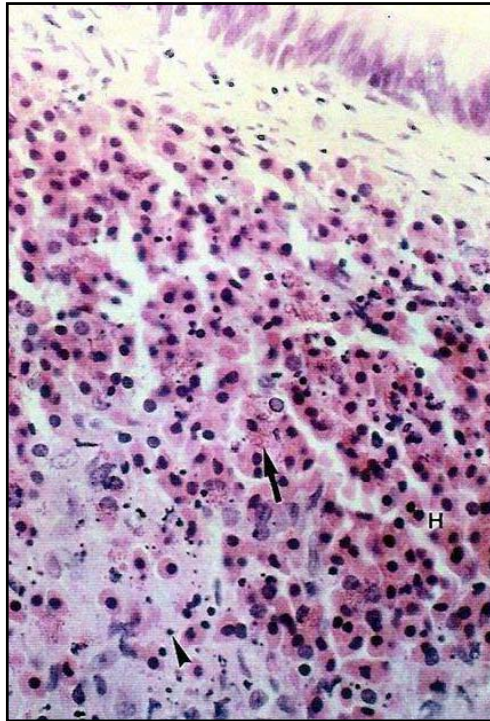
Hatchlings

- *Elseya latisternum* (5) 100% died 2xd10, 3xd20,
2 +ve for virus isolation
- *Emydura krefftii* (12) 42% died d16, 22, 24, 25, 29
3 +ve virus isolation
- No controls died or were positive for virus isolation

Challenge trials - histopathology

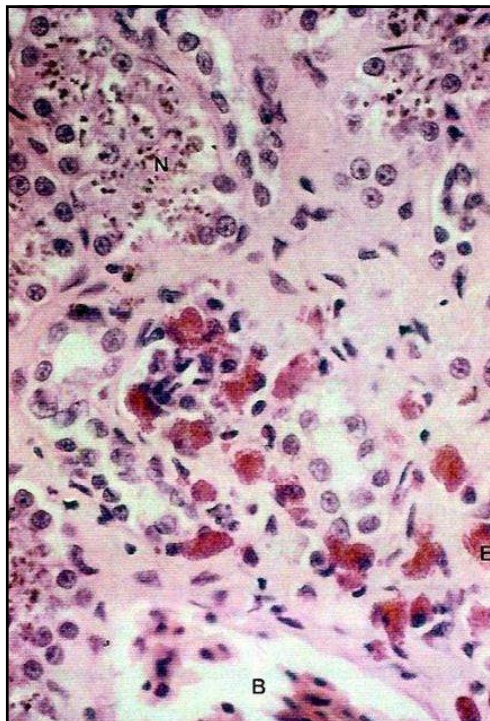
No obvious evidence of histological changes could be attributed to BIV infection in any of the challenged animals **APART** from turtle hatchlings.

Turtle hatchlings had lesions in the spleen, liver, kidney and submucosa of the gut.



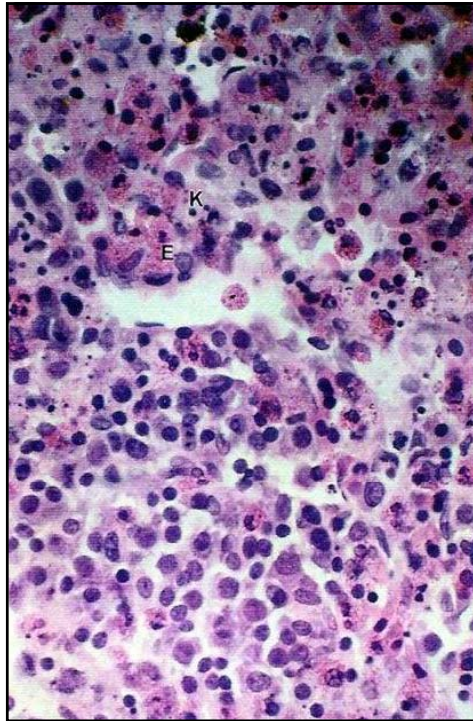
Submucosa

Haemorrhage, necrosis,
eosinophilic material
& karyorrhexis



Kidney

Necrosis and eosinophilia
near bloodvessel



Spleen

Eosinophilia and karyorrhexis
in necrotic area

Summary

Bohle iridovirus was found to be extremely virulent in hatchling tortoises (*Elseya latisternum* and *Emydura krefftii*), resulting in lesions in multiple organs and death (100 and 40% respectively).

Adult tortoises survived BIV-challenge and produced antigen-specific antibodies.

Thus, serological surveys of adult tortoises may be useful for determining the presence of BIV in northern Australia.

Current studies

More widespread serum survey beyond the advancing line of cane toads (*Bufo marinus*) that are invading the continent from East to West, to see if they are spreading the virus.

Immunological responses to ranavirus infection

Future directions

Sero surveys and challenge studies to enable risk analysis and potential impact to fauna from introduced ranavirus.