Chasing “PIX”

Sally Isberg and many, many more!

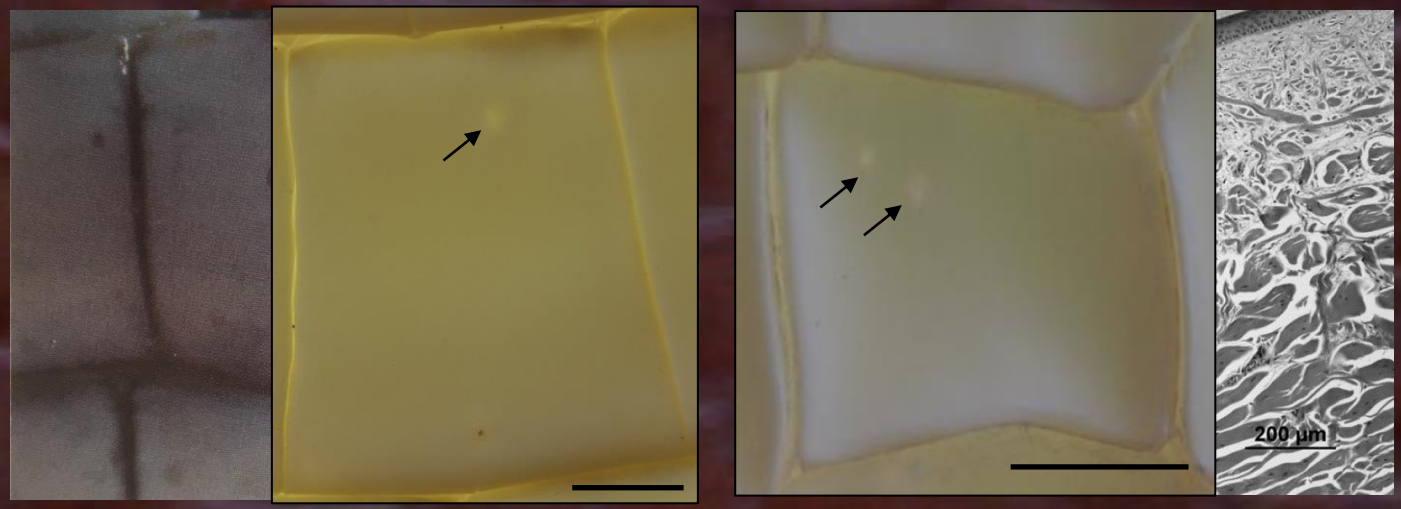
sally@crocresearch.com.au

Centre for Crocodile Research™

@CrocResearch

“PIX” defined

- <2 mm focal lesions
- Either singular or multiple (eg cloud pix, line pix,…)
- Normal or depressed contour
- Normal or abnormal keratin
- Name “pix” arose to describe the characteristic “ice pick” marks that occur on tanned skins from West Nile virus lesions.

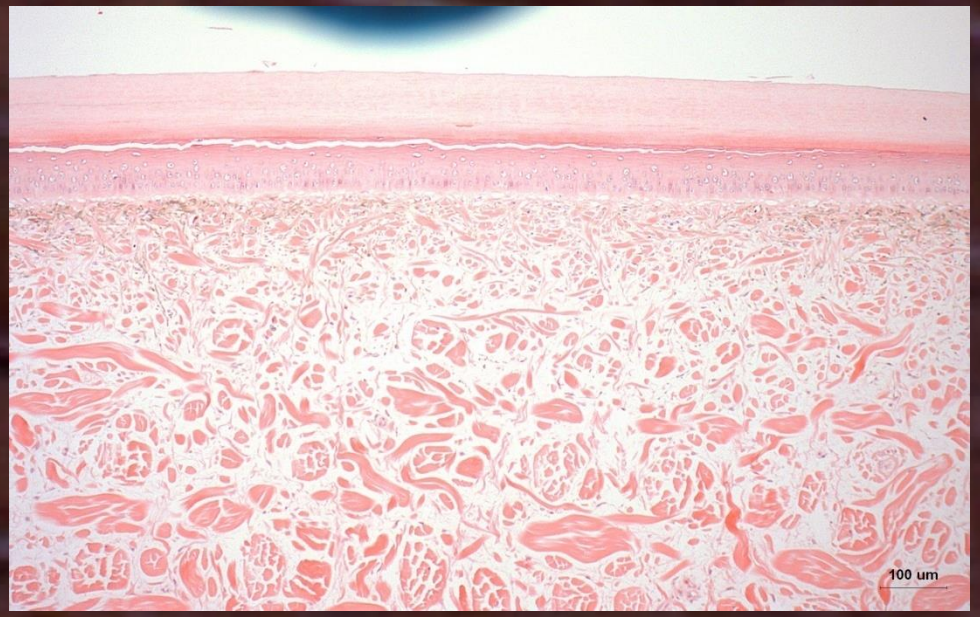


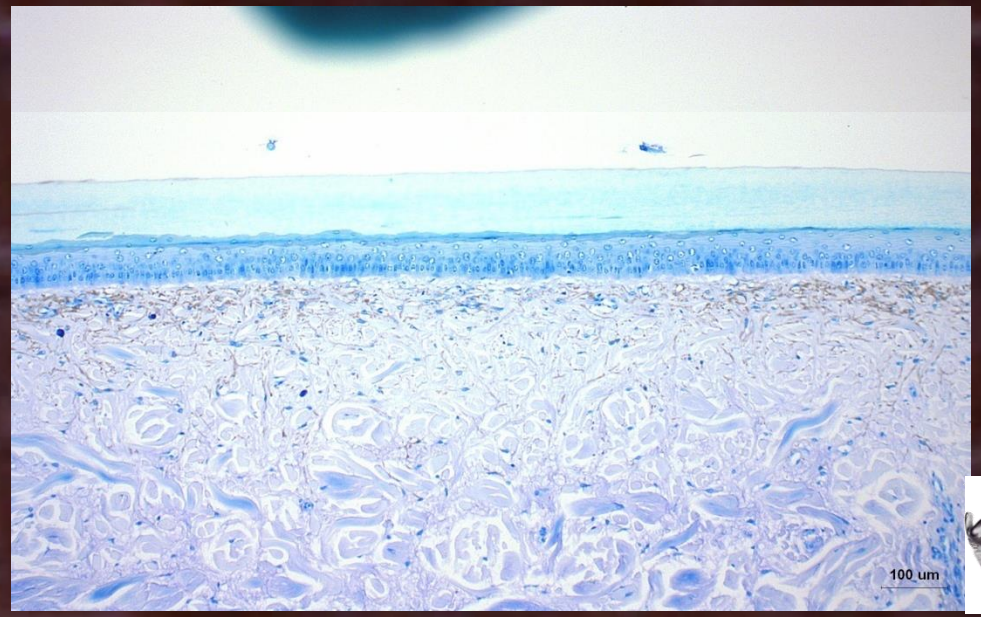
Navarez *et al*. (2008)

Centre for Crocodile Research

Tasked to find “PIX”

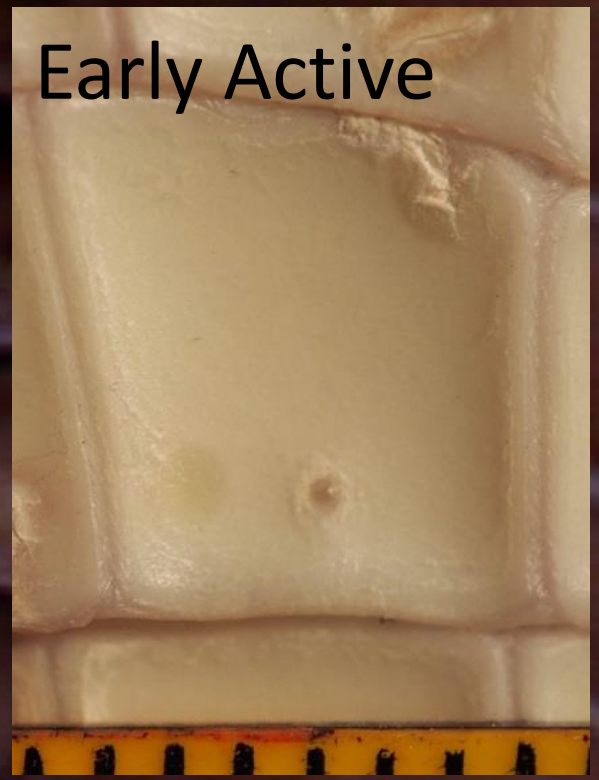
- In 2009, skin acceptance rates went from 80% acceptance to 80% rejection overnight.
- NT Minister asked Berrimah Veterinary Laboratory to be involved.
- Herein began the journey chasing pix.

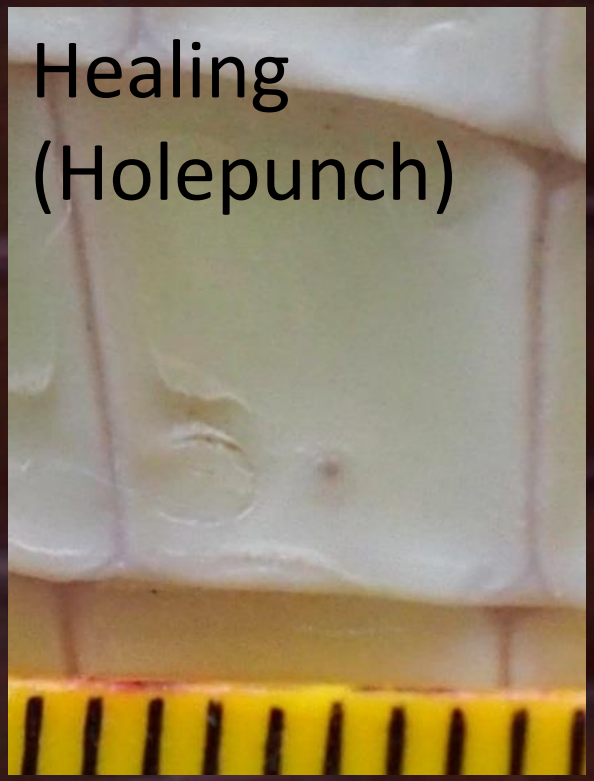




Centre for Crocodile Research

Gross poxvirus lesion stages

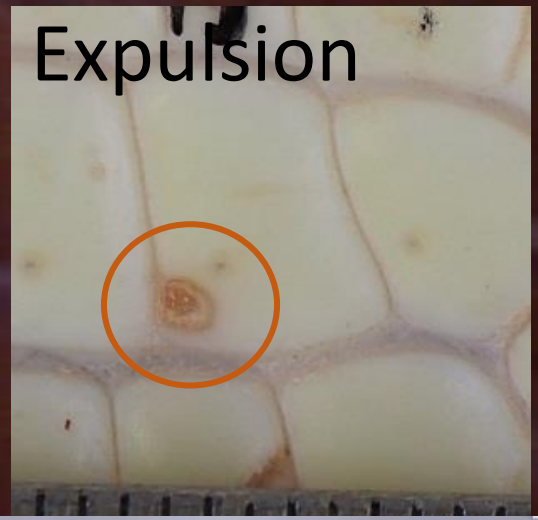


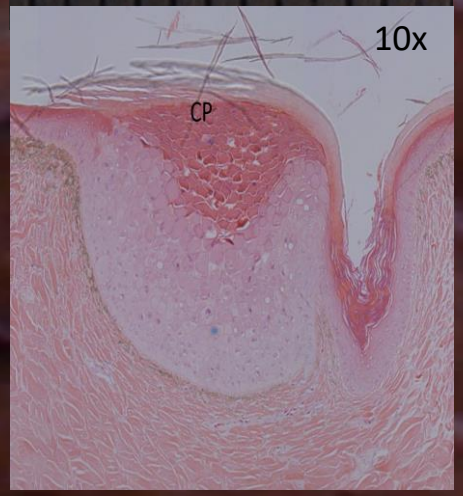


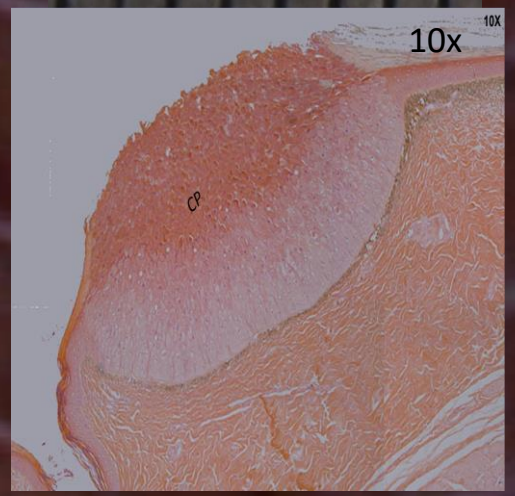
From Moore *et al.* (2017)

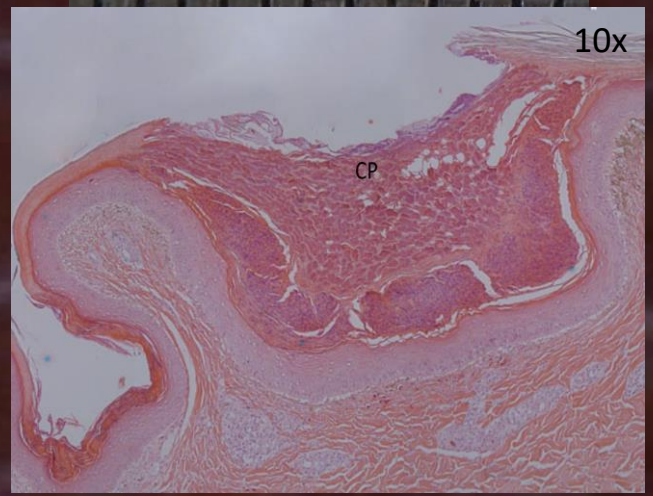
Centre for Crocodile Research

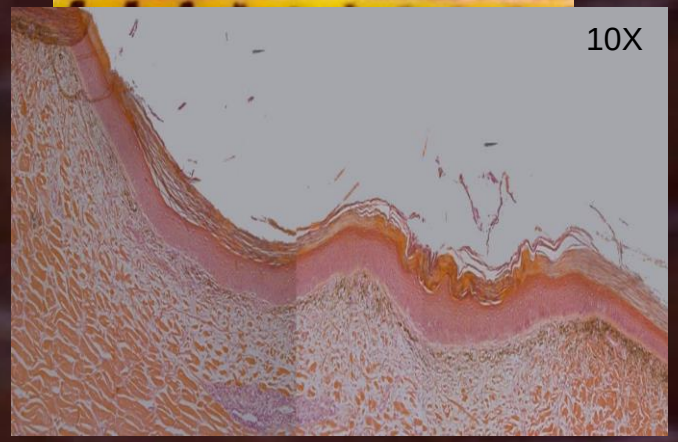
Histology of poxvirus lesion stages







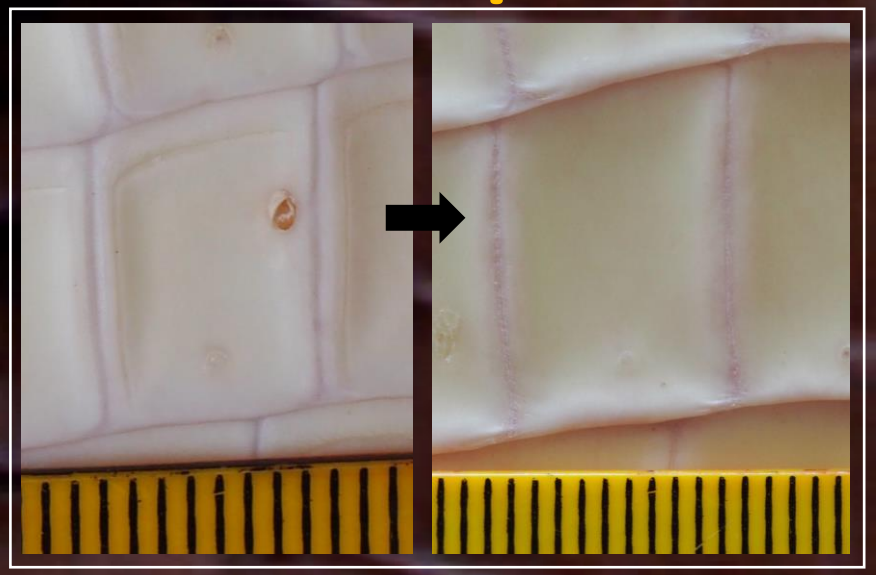


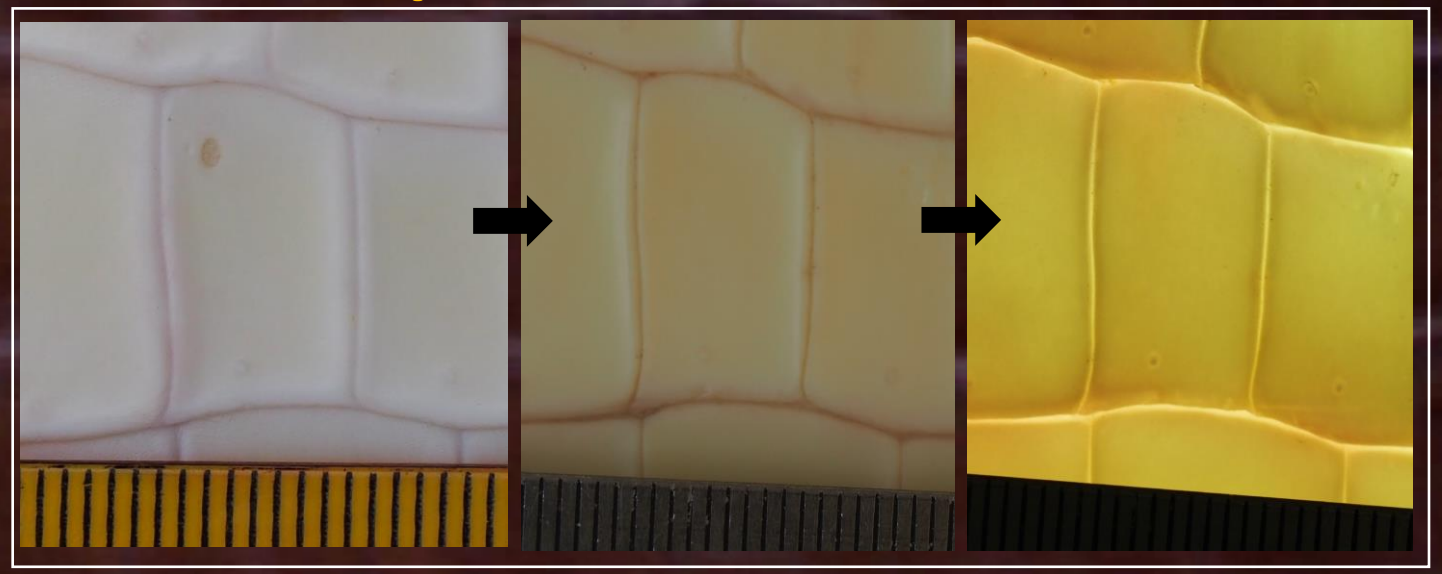


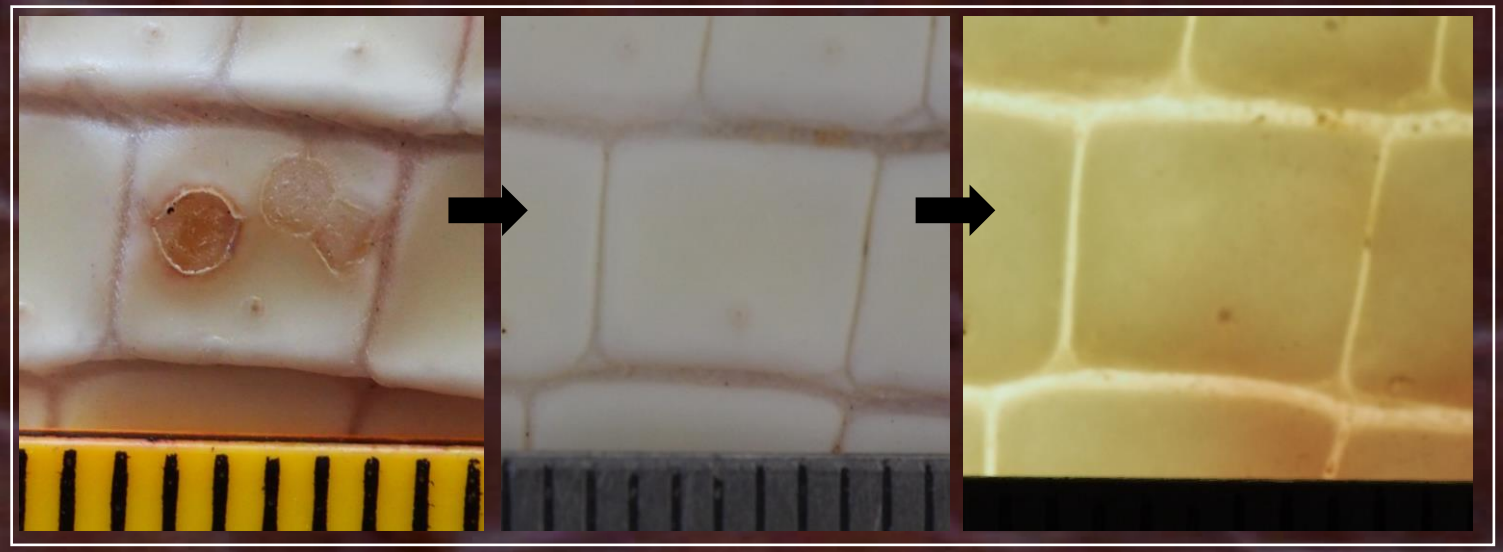
From Moore *et al.* (2017)

Centre for Crocodile Research

Examples of healed poxvirus lesions







From Moore *et al.* (2017)

Centre for Crocodile Research

Defining the limits of poxvirus lesions

Poxvirus

Dermatophilus

Herpesvirus

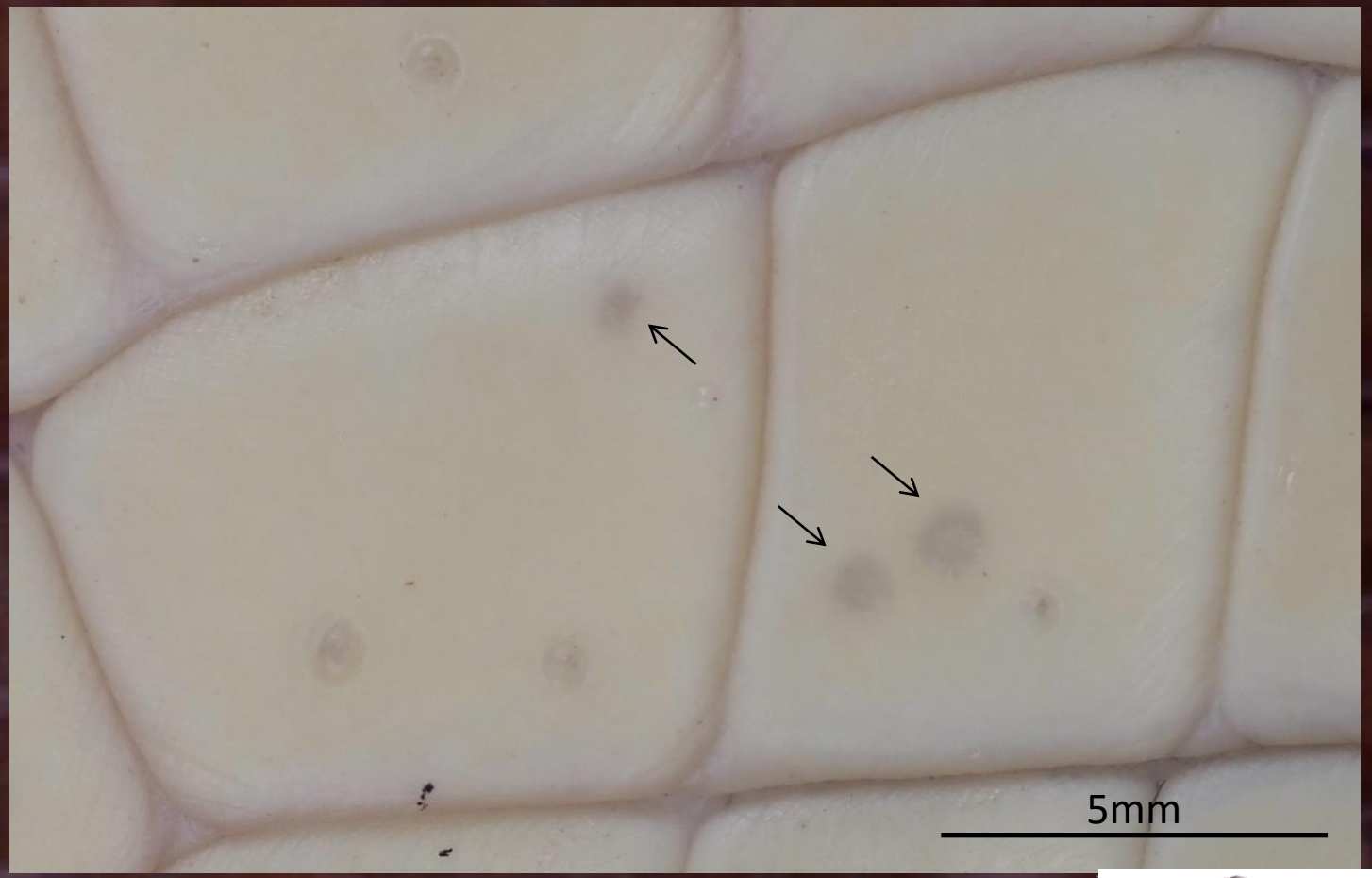
West Nile virus – Kunjin strain

Herpesvirus & WNV

Centre for Crocodile Research

Gross appearance of WNV lesions

- 1-2 mm
- No contour
- Normal keratin
- Active lesions are red-grey and obvious
- Older lesions are less obvious
- Lucent on the light table



Centre for Crocodile Research

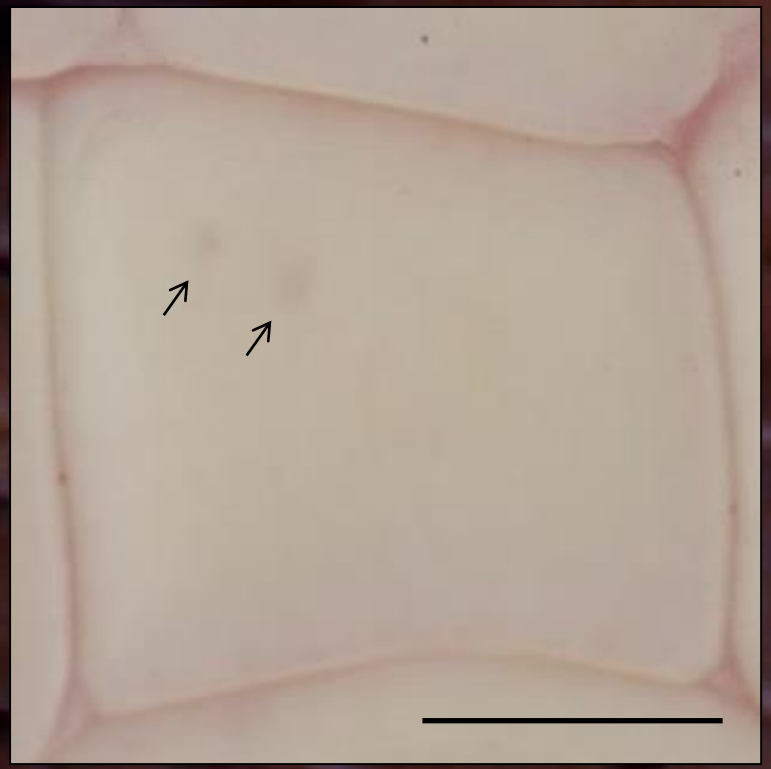
Gross and histopathology of WWD lesions

Light-table

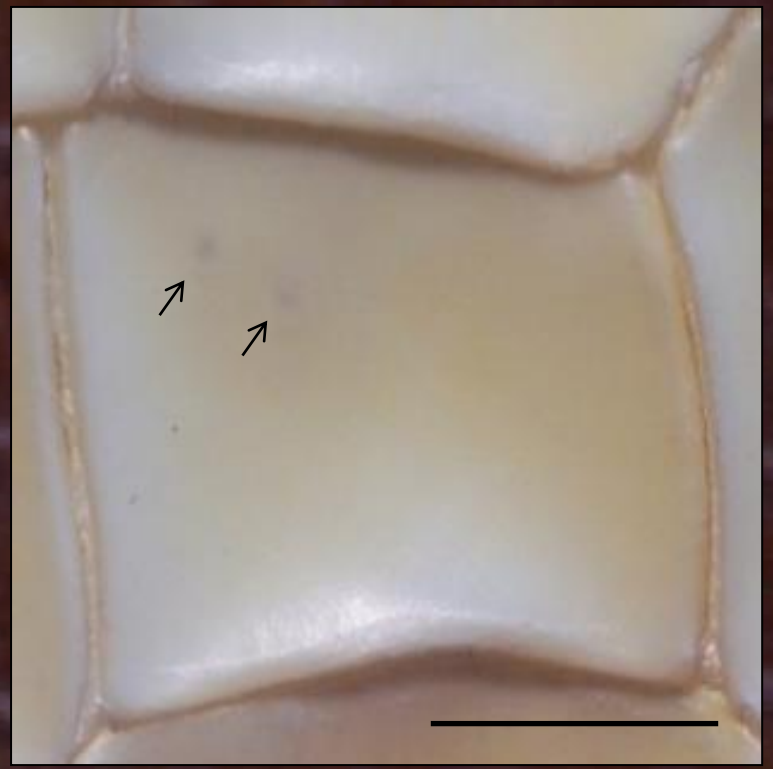
Centre for Crocodile Research

WNV time series

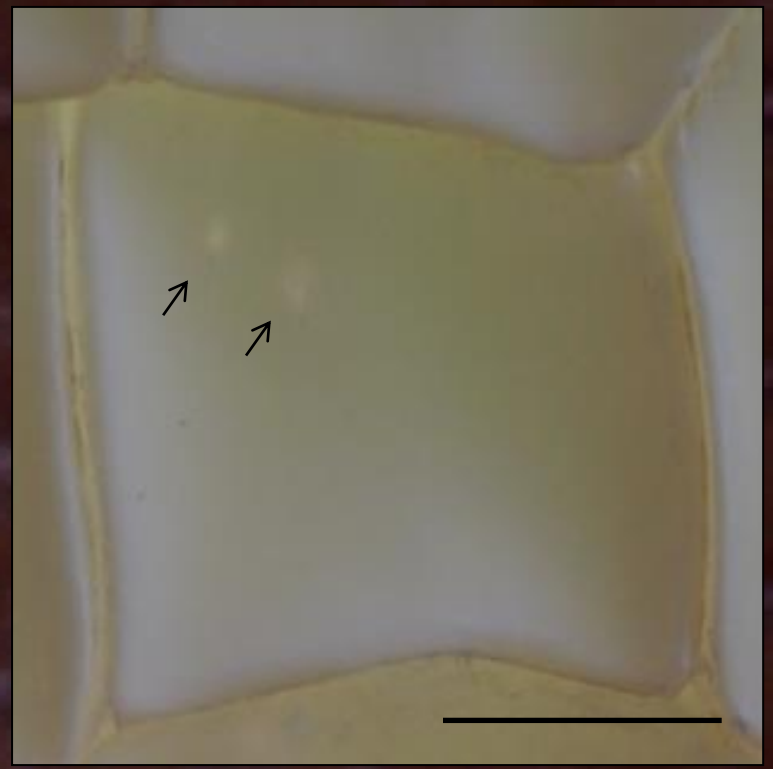
First identified



6 months later



Light-table

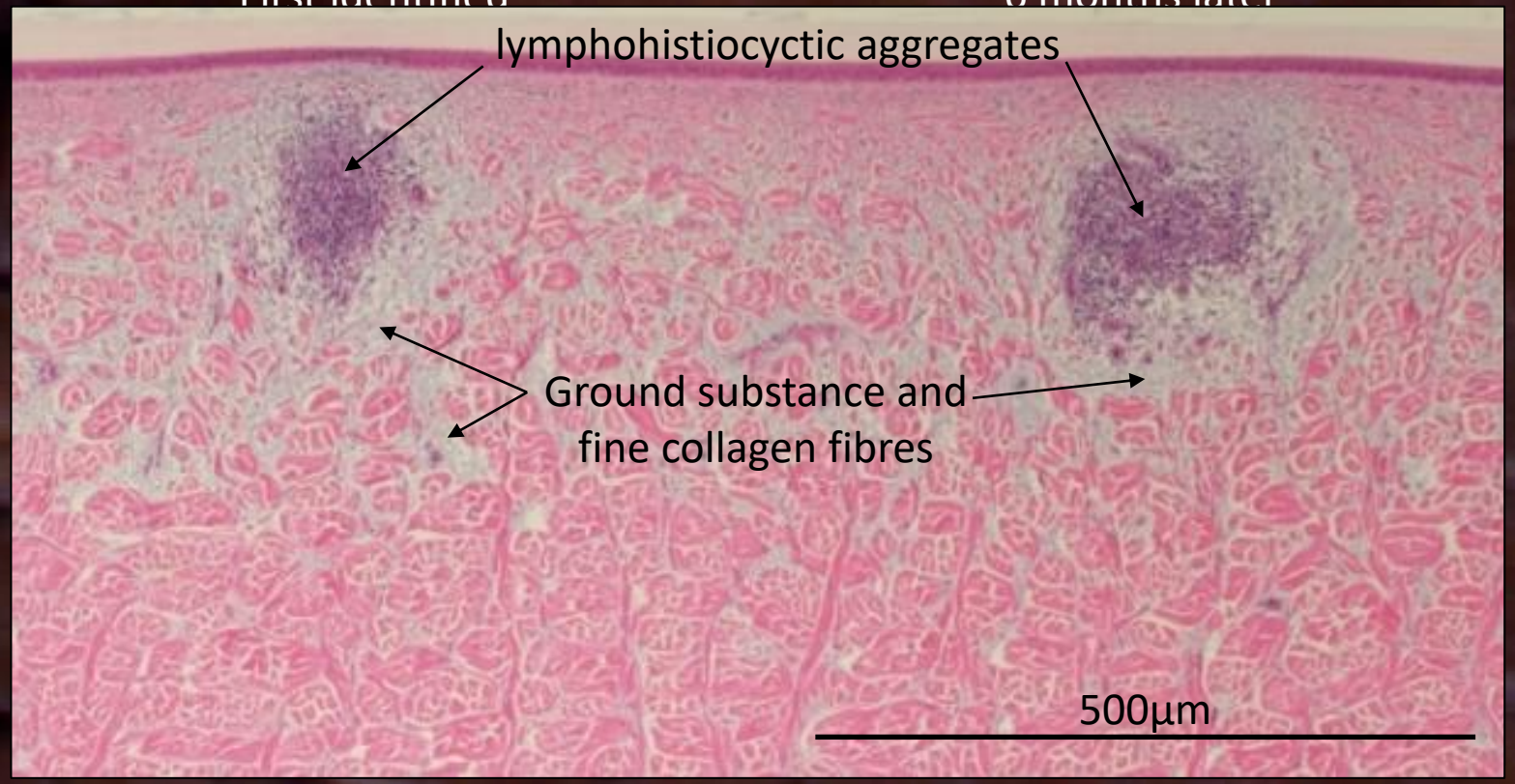


Centre for Crocodile Research

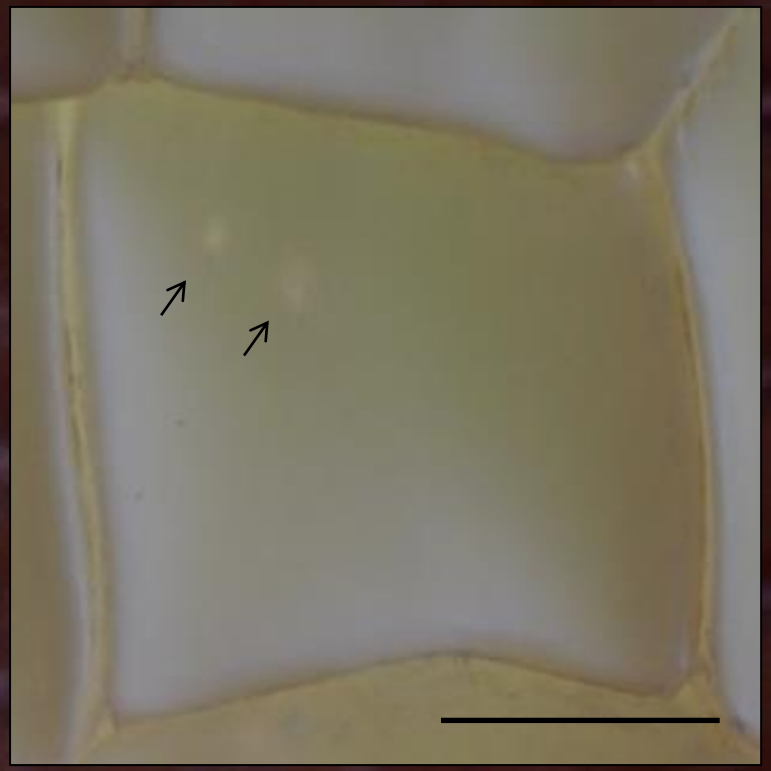
Histology of WSV lesions

First identified

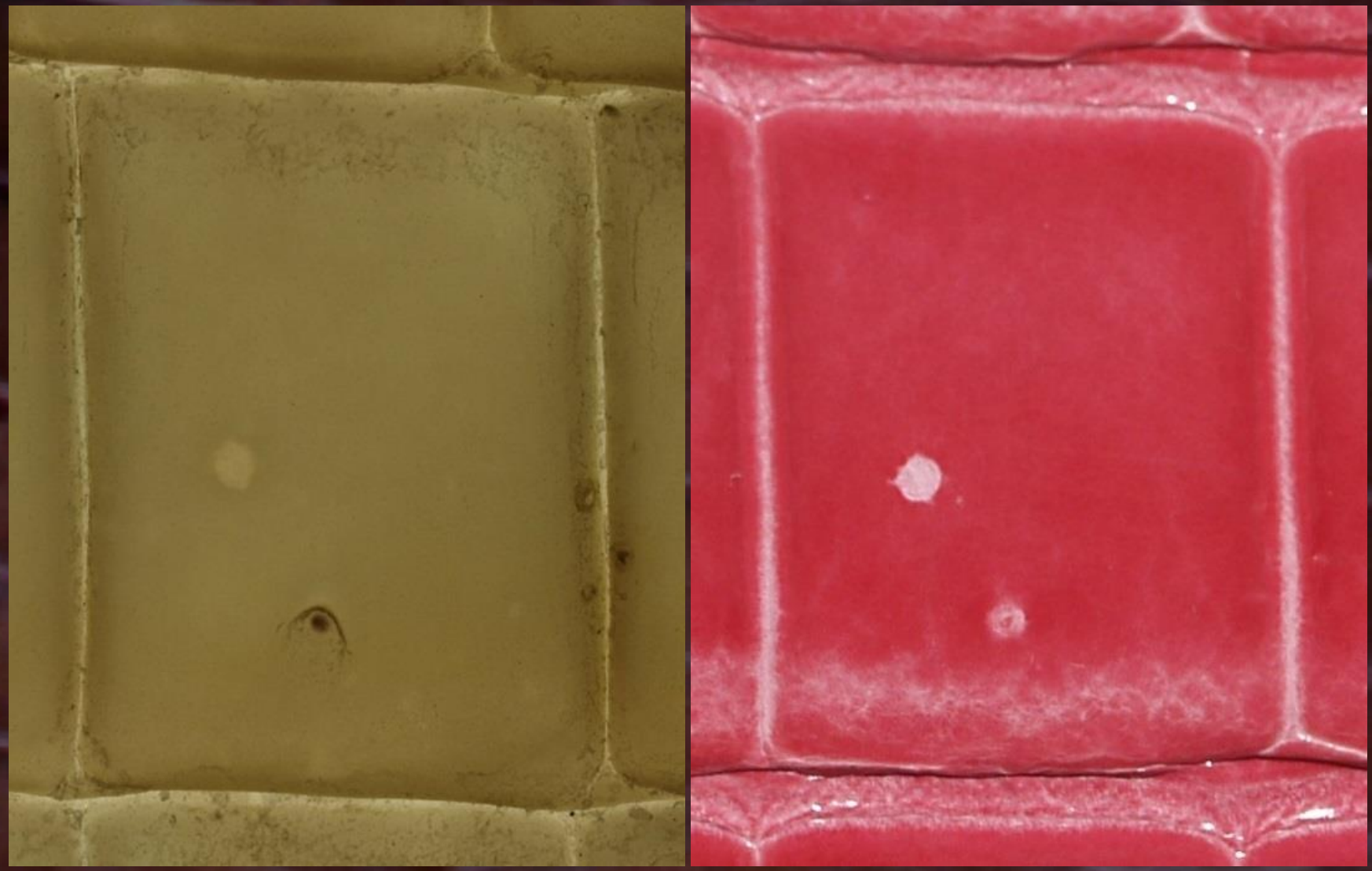
6 months later



Light-table



WNV = “PIX”



Centre for Crocodile Research

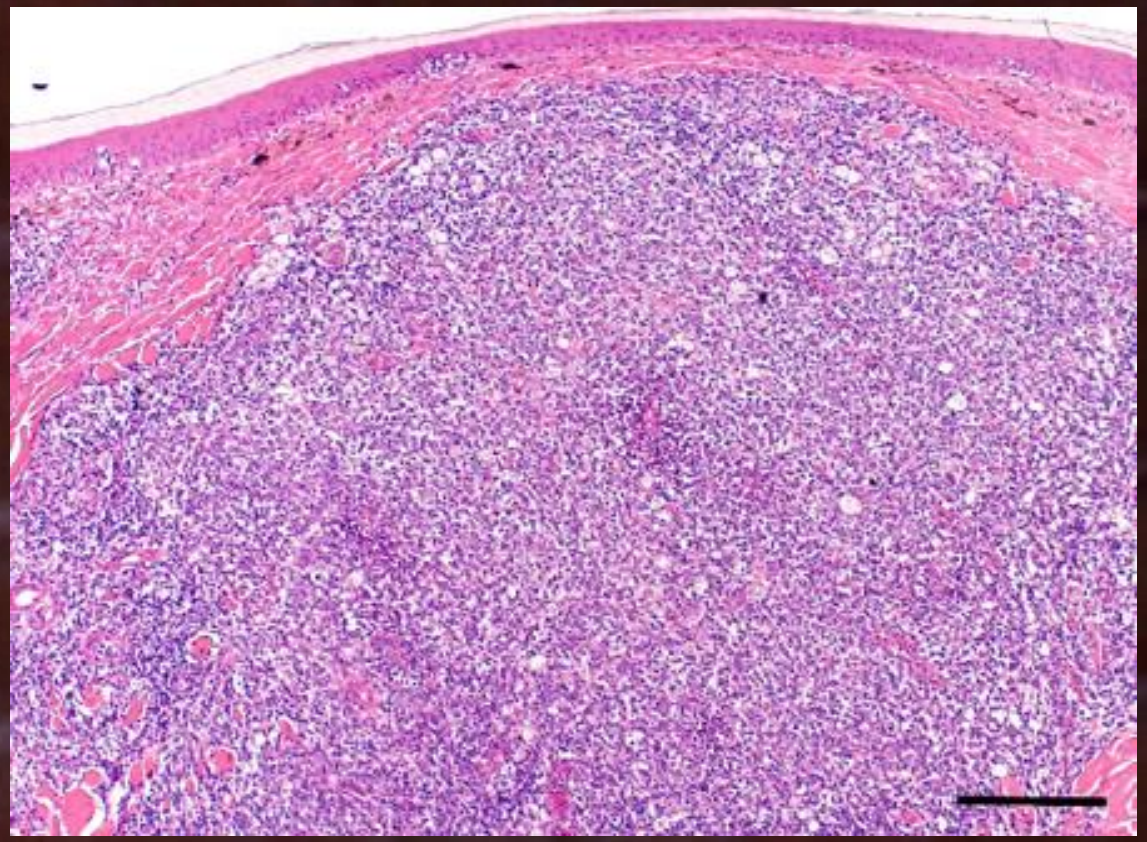
Gross appearance of Herpesvirus

- Lymphonodular skin lesions (LNS)
  - flank scales
  - belly scales.
- <5mm
- Normal - Raised contour
- Normal - Abnormal keratin
- Active lesions are red/grey and obvious

Centre for Crocodile Research

Histology of the herpesvirus

- Dermis infiltrated by large dense aggregates of lymphocytes
- The significant displacement of the dermis causes restructuring of the collagen
- Lucent on the light table

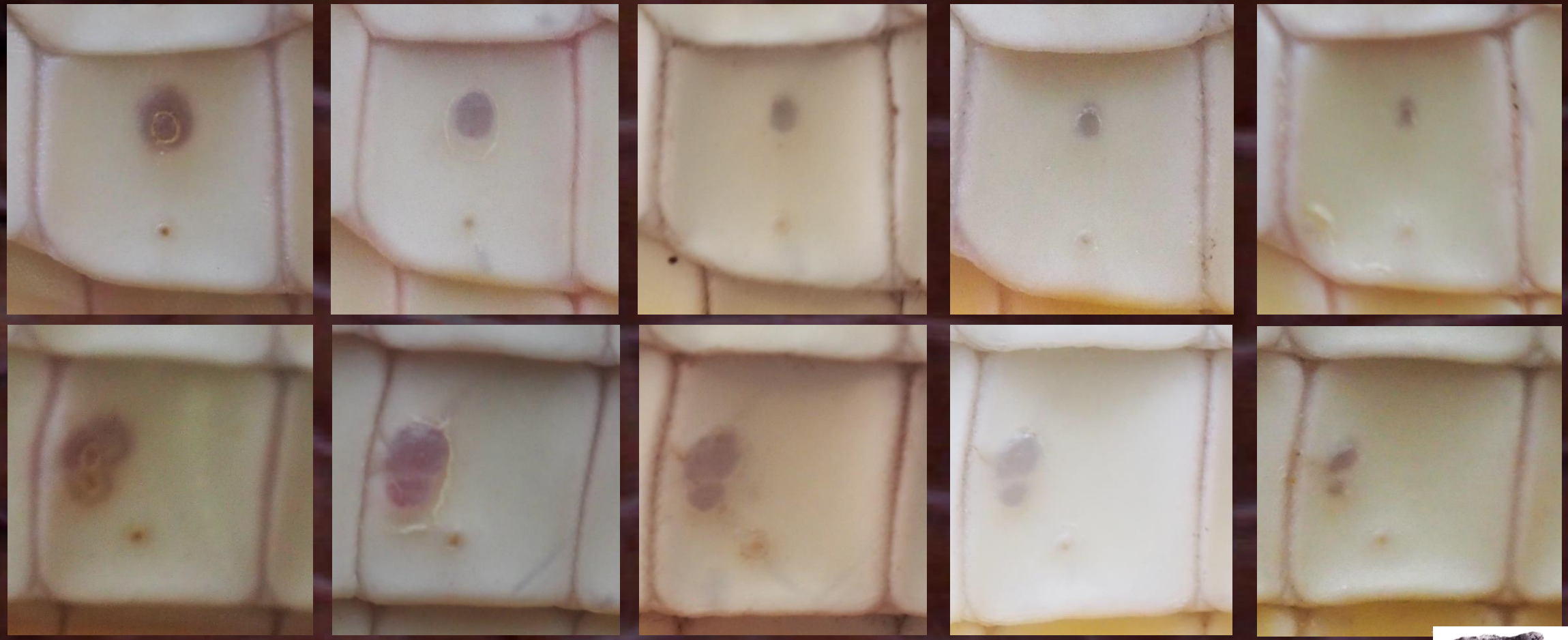


From Lott *et al*. (2018)

Centre for Crocodile Research

Herpesvirus time “Bites”

6 months



From Moore (2013)

Centre for Crocodile Research

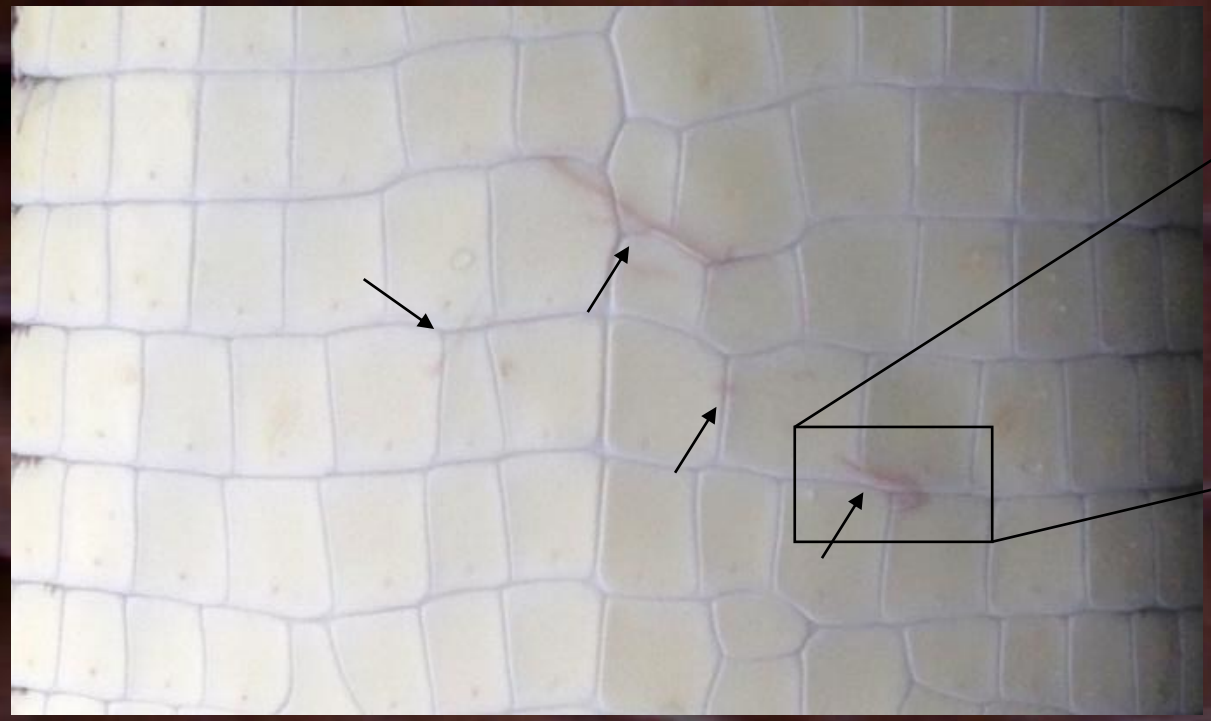
Dermatophilus

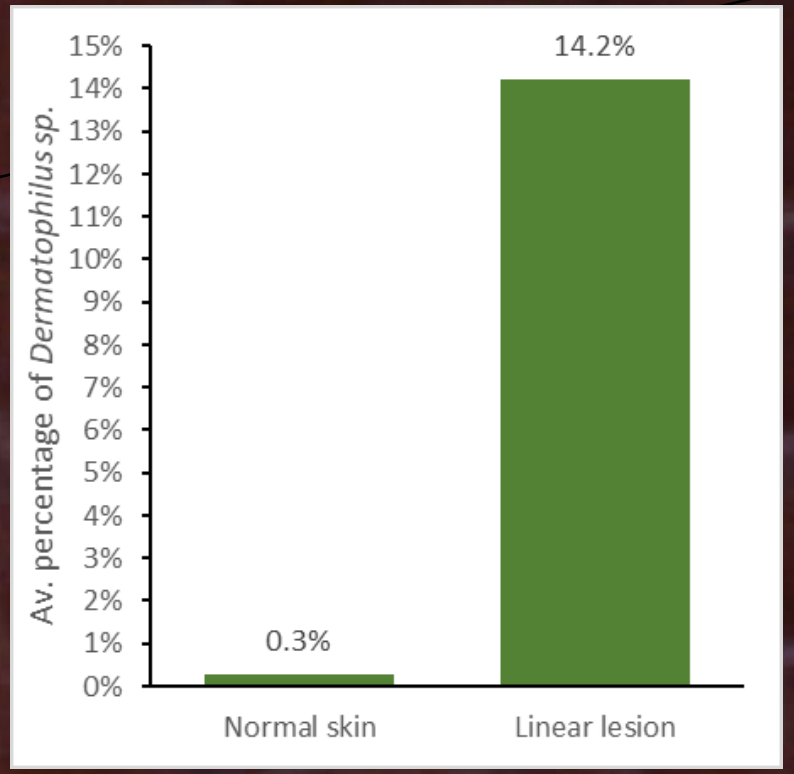
- First defined to be responsible for the condition dermatophilosis or ‘brown spot’ disease.
- Brown spot lesions are characterized by:
  - brown or red lesions;
  - abnormal keratin; and
  - 1-4 mm spots → 2 cm^2 ulcerations → 5cm linear erosions.
- Located predominantly over alpha-keratin regions (ISOs and hinges) on the abdomen, tail and head.

From Lott *et al*. (2018)

Centre for Crocodile Research

Dermatophilus in linear lesions

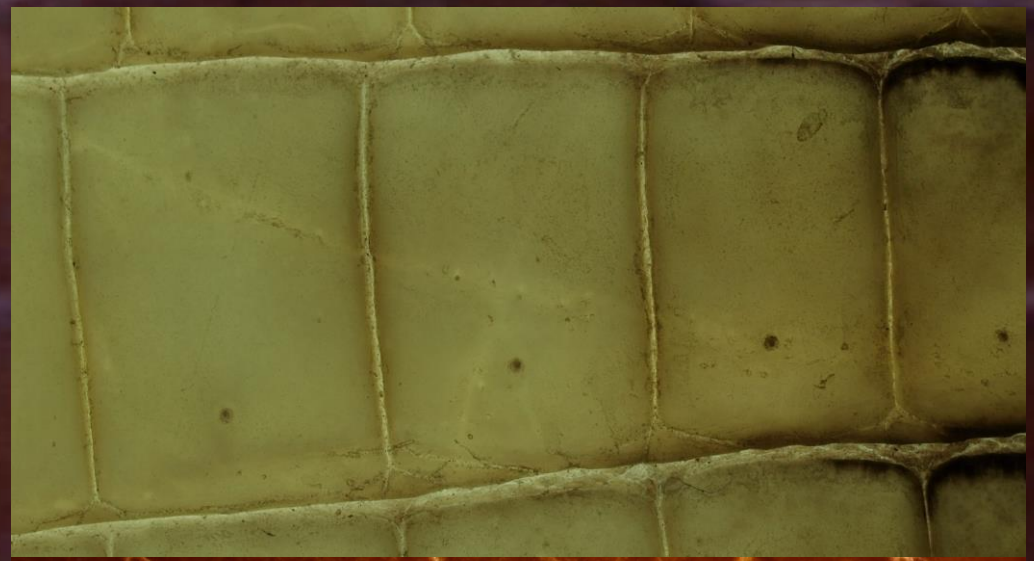


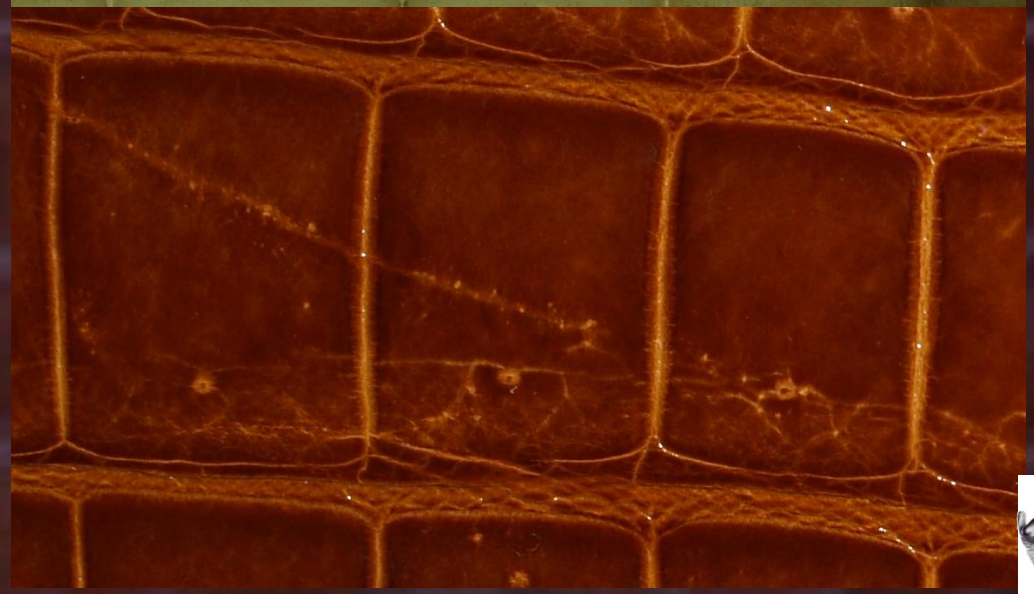


From Isberg *et al*. (2019)

Centre for Crocodile Research

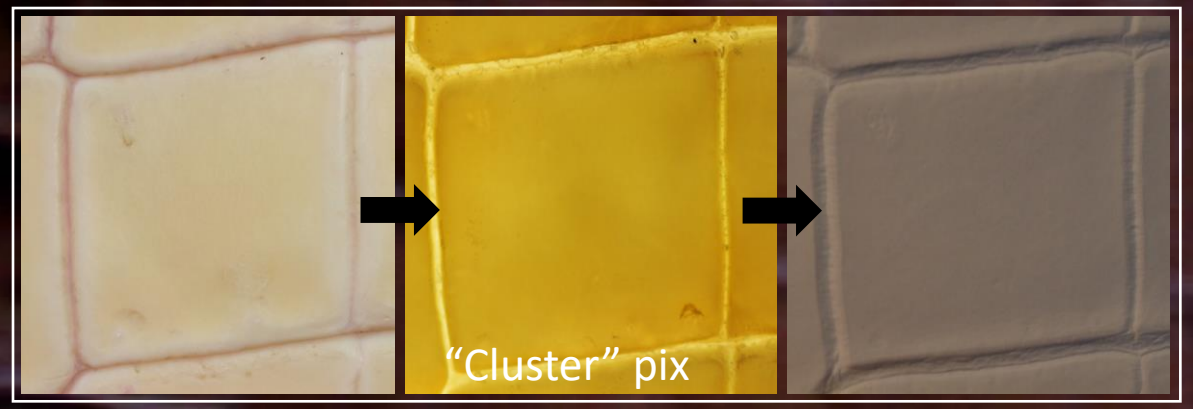
Dermatophilus ≈ line pix





Centre for Crocodile Research

Bacterial pix



All of these lesions developed whilst in finishing pens!

Centre for Crocodile Research

“Pix” re-defined

- Collective term for any <2mm focal lesion that is lucent on the light-table; not just WNV as originally described.
- Their characteristics (keratin, contour) can be indicative of causation.
- How do determine what is causing your “pix”?

Centre for Crocodile Research

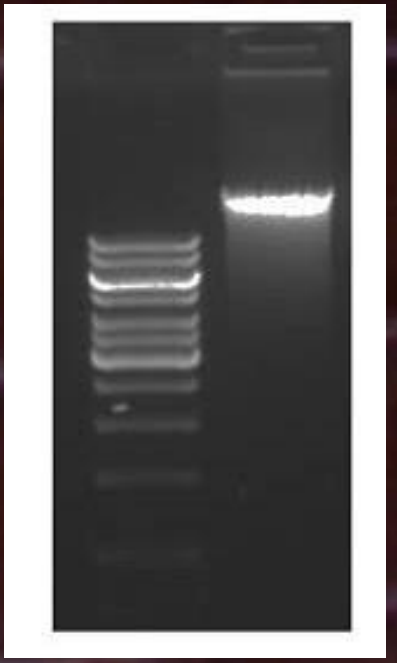
How do determine what is causing your “pix”?

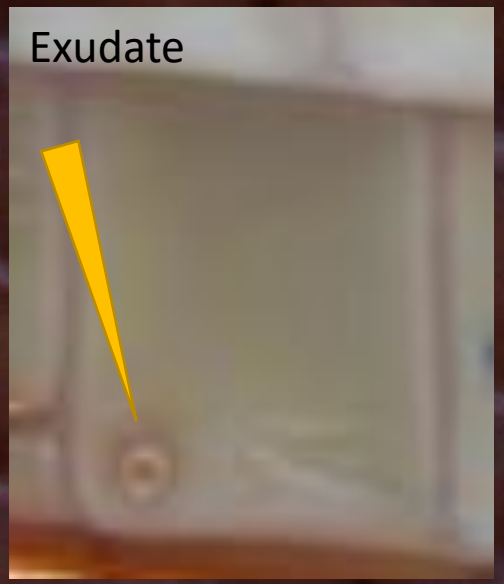
1. Do my Crocodilian Skin in 3D course!
2. Standardise the terminology within your team!
   - WNV pix
   - Bacterial pix
   - Line pix
   - Thumbnail
   - Cloud pix
   - Herpesvirus
   - PITS (very different from “pix”)
3. Determine what testing you require based on the above.

Centre for Crocodile Research

Diagnostics of the lesions

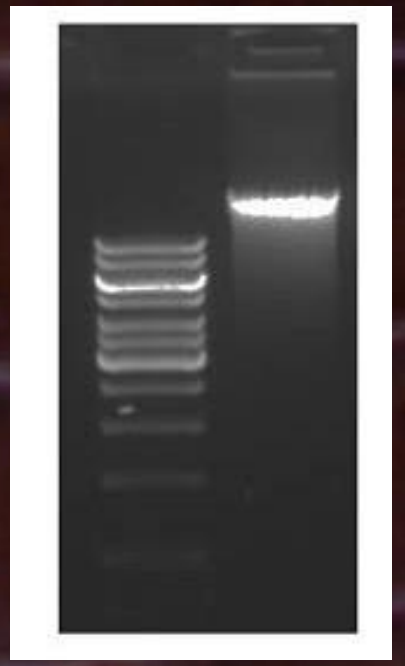
- Microbiology
- Genetic testing – specific PCRs
  or 16S bacterial sequencing



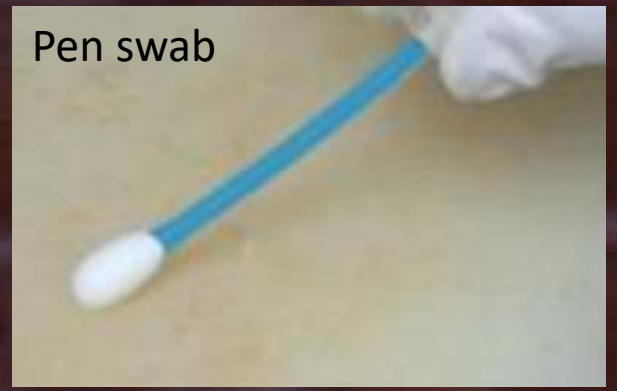


Centre for Crocodile Research

Diagnostics of the environment



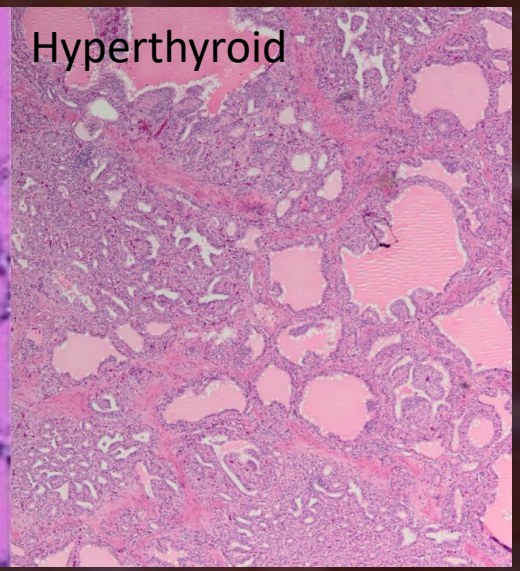
- Microbiology
- Genetic testing – specific PCRs
  or 16S bacterial sequencing
- Mosquitoes



Centre for Crocodile Research

Animal diagnostics

- Functioning thyroids – histology and hormone levels (ELISA)
- Antibody detection – serology (VNT, ELISAs)



Centre for Crocodile Research

Prevention

- Dependent on causative agents:
- WNV –
    - Biosecurity - bird and mosquito control and deterrents
    - Water monitoring
- Herpesvirus –
    - Water monitoring
    - Hygiene
    - Stress reduction
- Dermatophilus/bacterial pix/cluster pix/cloud pix
    - Water quality
    - Hygiene protocols

Centre for Crocodile Research

Managing water quality

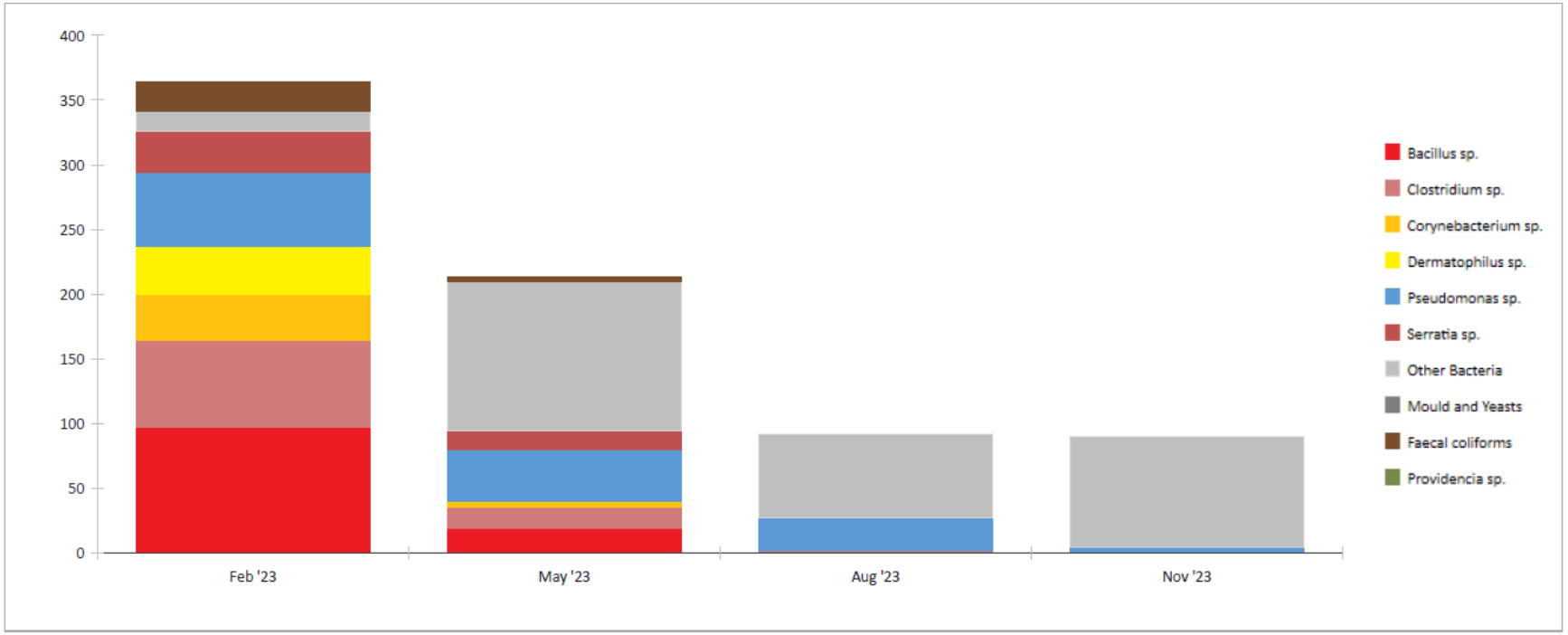
Centre for Crocodile Research
PO Box 329,
NOONAMAH NT 0837
Phone: 0427 427 489
Email: sally@crocresearch.com.au
@CrocResearch
@IsbergSally
Centre for Crocodile Research

Farm	Puk Puk Farming Inc
Area	Finishing Pen
Pen Name	ZHO77
Test Date	18/08/2023

Important skin quality indicator		
Bacillus sp.	0.0	cfu/ml
Clostridium sp.	2.0	cfu/ml
Corynebacterium sp.	0.0	cfu/ml
Dermatophilus sp.	0.0	cfu/ml
Serratia sp.	0.0	cfu/ml
Commensal		
Pseudomonas sp.	25	cfu/ml

Health/water quality indicators		
Faecal indicators	0	cfu/ml
Providencia sp.	0	cfu/ml
Total counts		
Bacteria	92	cfu/ml
Moulds	4	cfu/ml
Yeasts	6	cfu/ml
Total overall count	102	cfu/ml

Test Date	Since Water Change	Chemical Used	Free Cl	Last PP	Pen
Aug 2023	19 days prior	Chlorfoam	0	16/07/2023	ZHO77
Jul 2023	-	-	-	-	-
Jun 2023	-	-	-	-	-
May 2023	-	-	0	-	-
Aug 2022	-	-	-	-	-



Centre for Crocodile Research

Summary

- Understand what the causative agents of your skin quality issues are.
- Develop and implement a strategic plan to reduce the prevalence of these organisms.
- Continue to monitor and assess these organisms through proactive monitoring strategies.
  - More frequent sampling in beginning.
  - Subsiding to less frequent maintenance checks once stabilized.
- We will continue to find new causations – no complacency!

Centre for Crocodile Research

Chasing “PIX”

Sally Isberg and many, many more!

sally@crocresearch.com.au

Centre for Crocodile Research™

@CrocResearch