



Pathological and immunological consequences of different blood sampling procedures in mice or Blood sampling in mice: Do we have “The Best Method”?

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The best way of blood sampling mice

Research Question:

Which method is the best when taking a blood sample in mice?

Short answer:

Long answer:



6 methods – 3 assessment points

Histopathology

Lesions,
inflammation and
healing

Markers of Inflammation

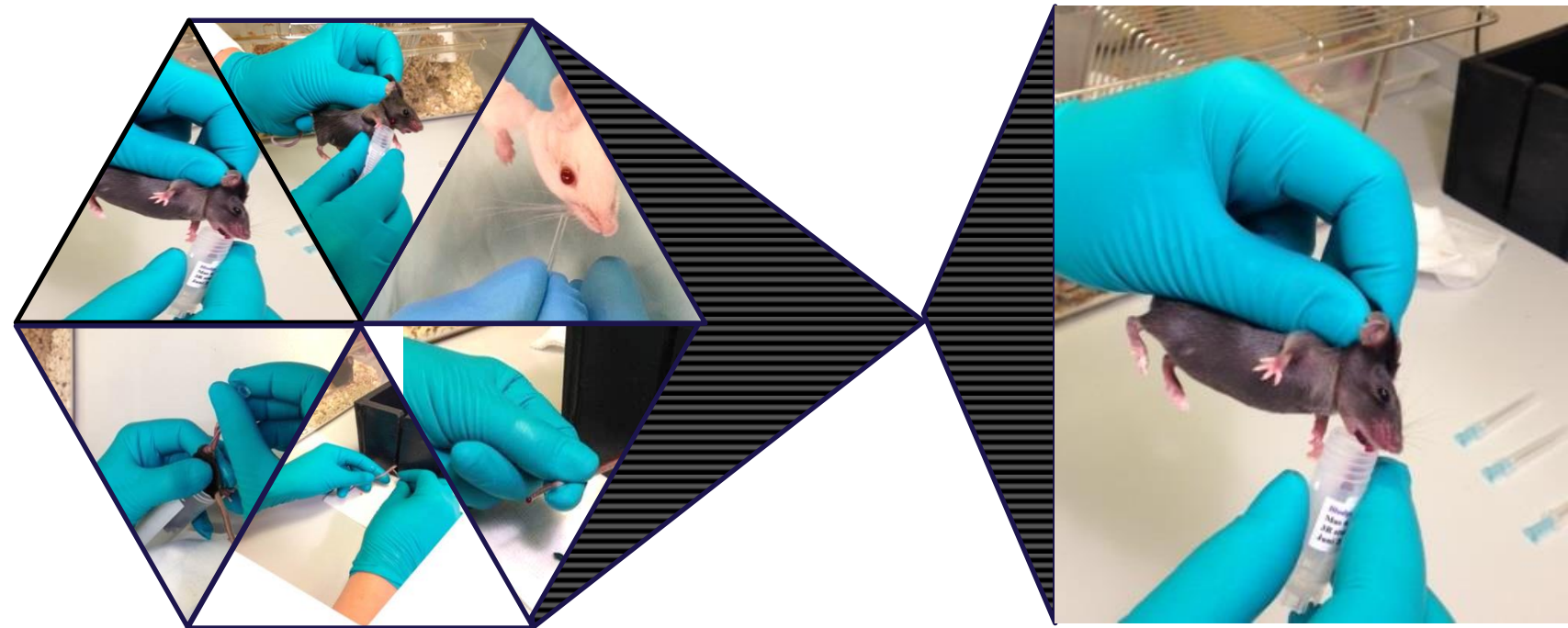
Expression of
inflammatory genes

Animal Welfare

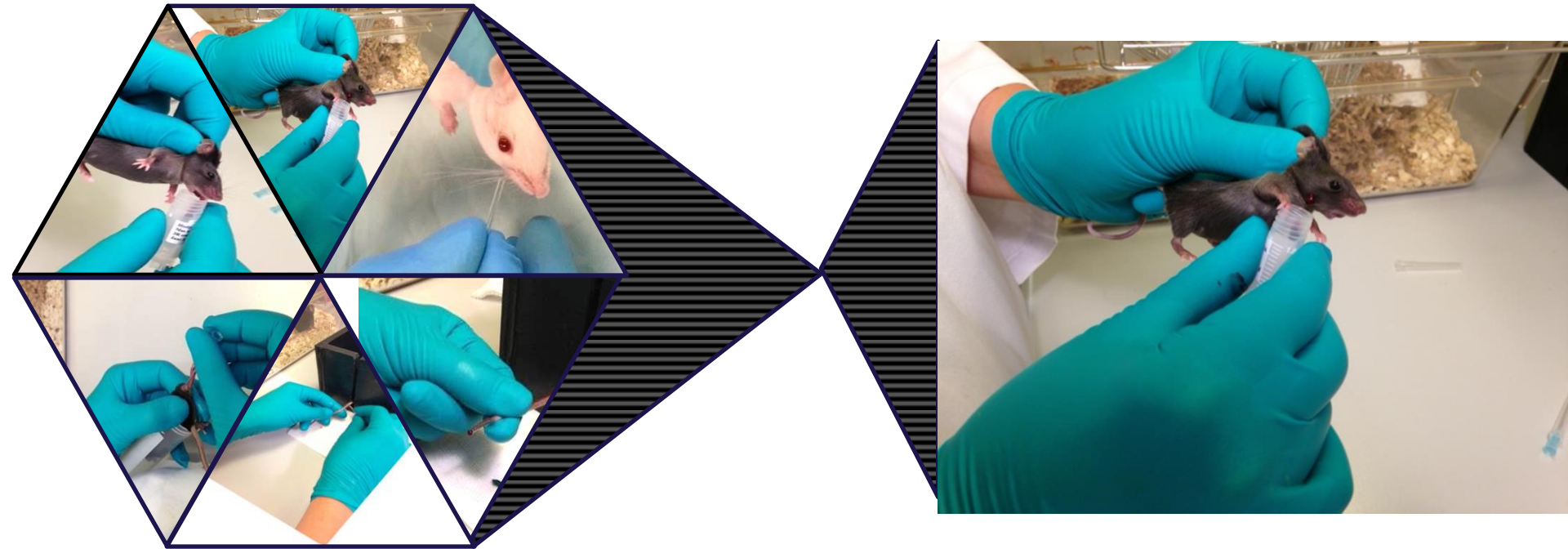
Stress, appetite and
body weight



6 methods – 3 assessment points



6 methods – 3 assessment points



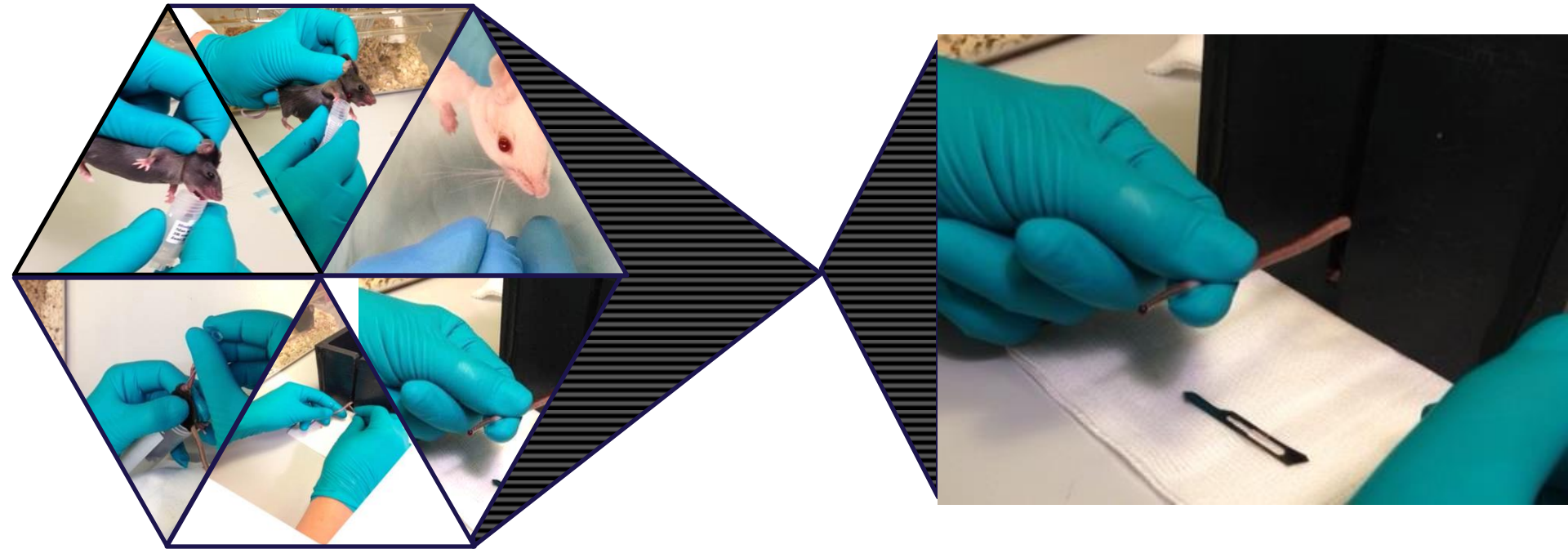
6 methods – 3 assessment points



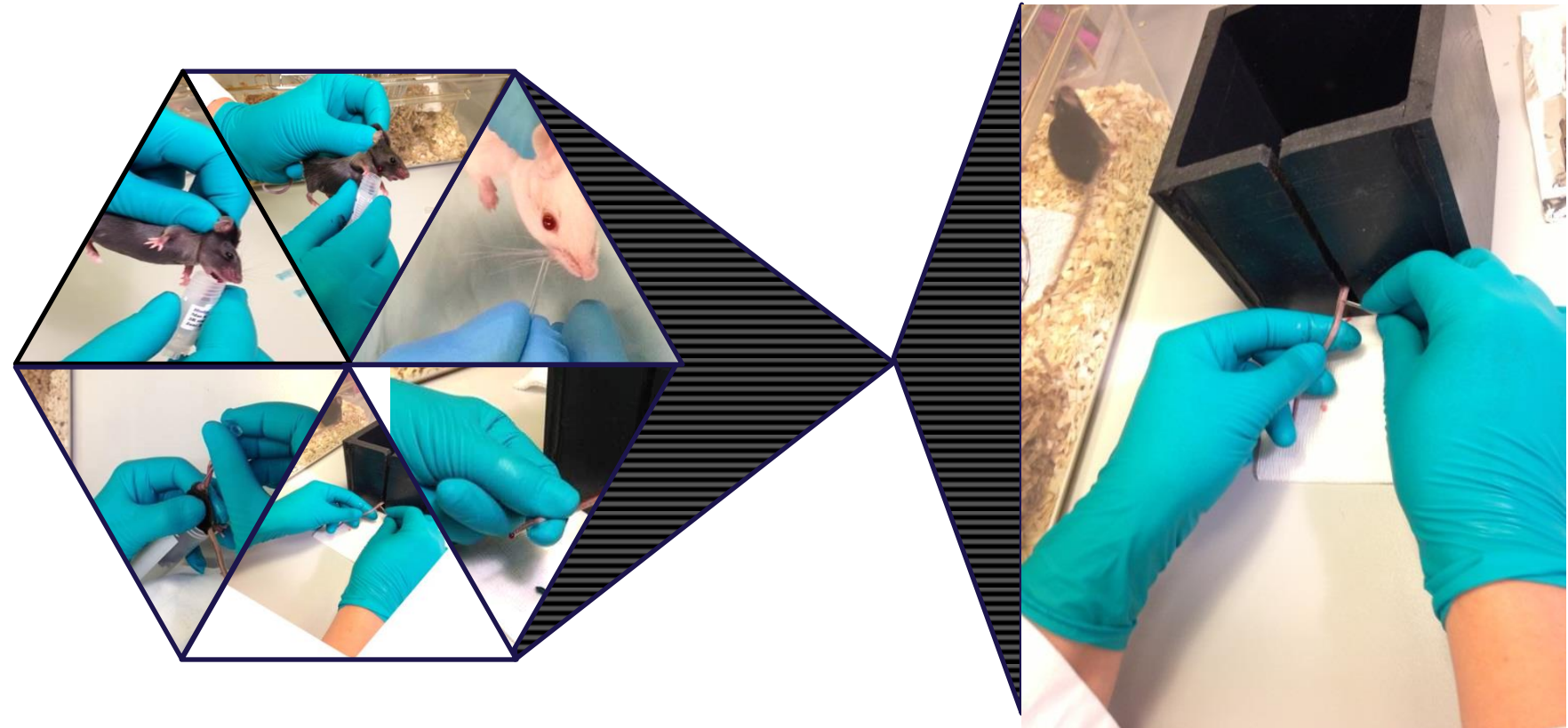
6 methods – 3 assessment points



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Histopathology

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Number of mice in each group shown as nHist (number of mice for histopathology) + nImm (number of mice for immunology)

Time for euthanasia after blood sampling	Blood Sampling Method							nHist + n Imm
	Tail	TTip	Eye	Tongue	Cheek	VSaph	Control	
T0 - Baseline	-	-	-	-	-	-	2 + 2	4
T1 - 6h	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	4 (iso)	28
T2 – 12 h	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	-	24
T3 – 24 h	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	4 (iso)	28
T4 – 2 days	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	-	24
T5 – 4 days	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	-	24
T6 – 6 days	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	-	24
T7 – 8 days	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	6 cort	30
T8 – 10 days	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	-	24
T9 – 12 days	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2	2 + 2		24
Total	18 + 18	18 + 18	18 + 18	18 + 18	18 + 18	18 + 18	18	234



Summary of histopathological findings

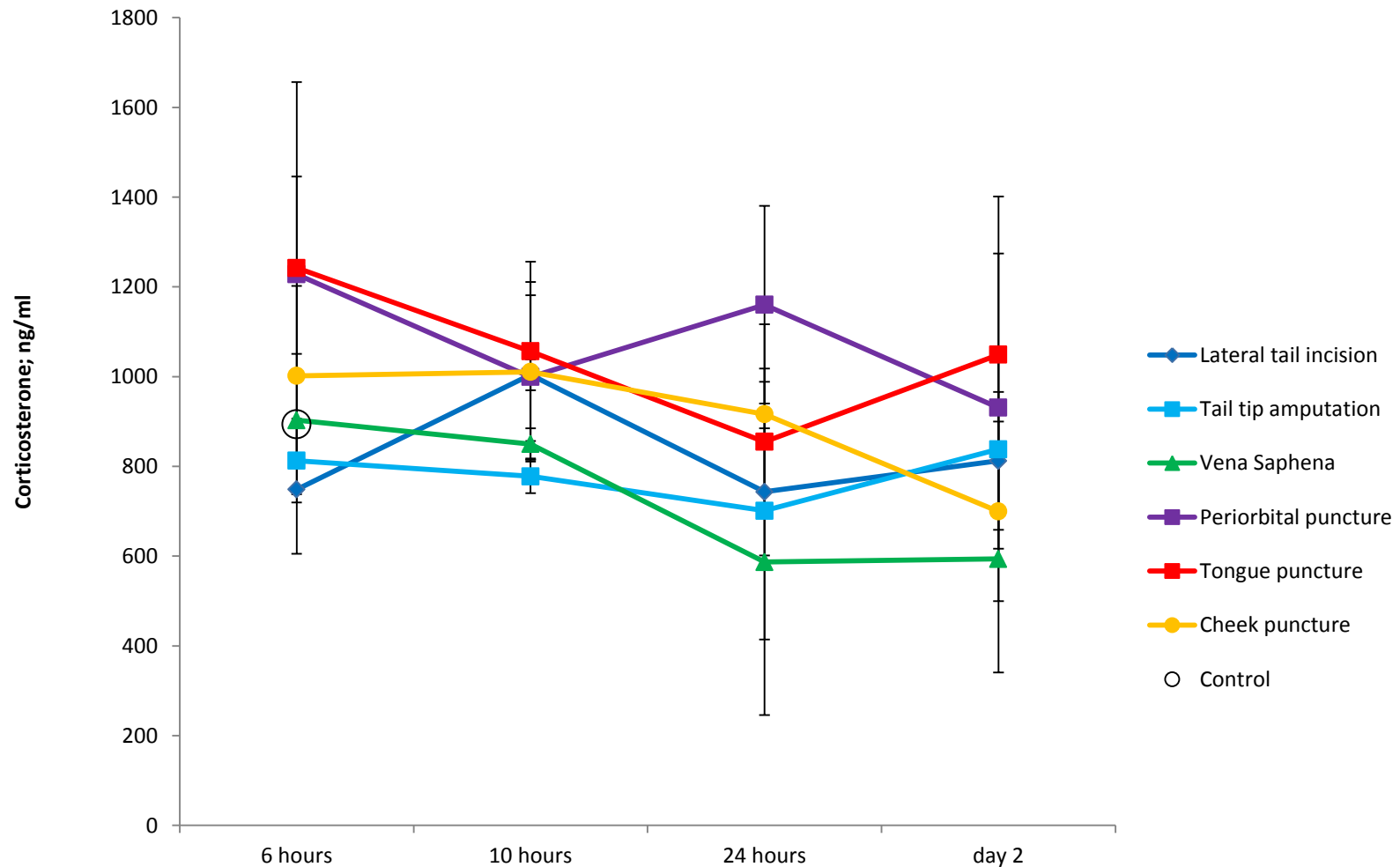
Histopathological finding	Blood Sampling Method				
	Tail	TTip	Tongue	Cheek	Eye
Completion of migration of epithelial skin cells (wound healing)	Day 4	No complete wound healing on day 12	N/A	Day 4	N/A
Acute inflammation (infiltration dom. by neutrophil cells)	Up to day 4	Up to day 6	Up to day 1	Up to day 2	Up to day 1
Micro abscesses	No	No	Yes (day 2 +8)	No	No
Trichogranuloma	No	No	Yes	Yes	Yes
Muscular necrosis and regeneration	Yes	No	Yes	Yes	Yes
Evidence of tissue trauma on day 12	Yes	Yes	Yes	No ID	Yes

Summary of immunological factors

Immunology	Blood Sampling Method					
	Tail	TTip	Vena Saphena	Tongue	Cheek	Eye
Systemic inflammation						
Haptoglobin (ELISA)	***	-	***	-	-	**
Gen expression						
S100A8/A9	x 30	x 30	x 100	-	x 200	-
Cxcl2	x 1100	x 1000	x 1000	-	x 11	x 100
IL-6	x 1000	x 11	x 100	-	x 10	x 100
IL-1b	x 100	x 3000	x 300	-	x 13	x 100
IL-33	x 6	x 4	-	-	-	-
Nlrp3	x 70	x 30	x 30	-	x 5	x 10

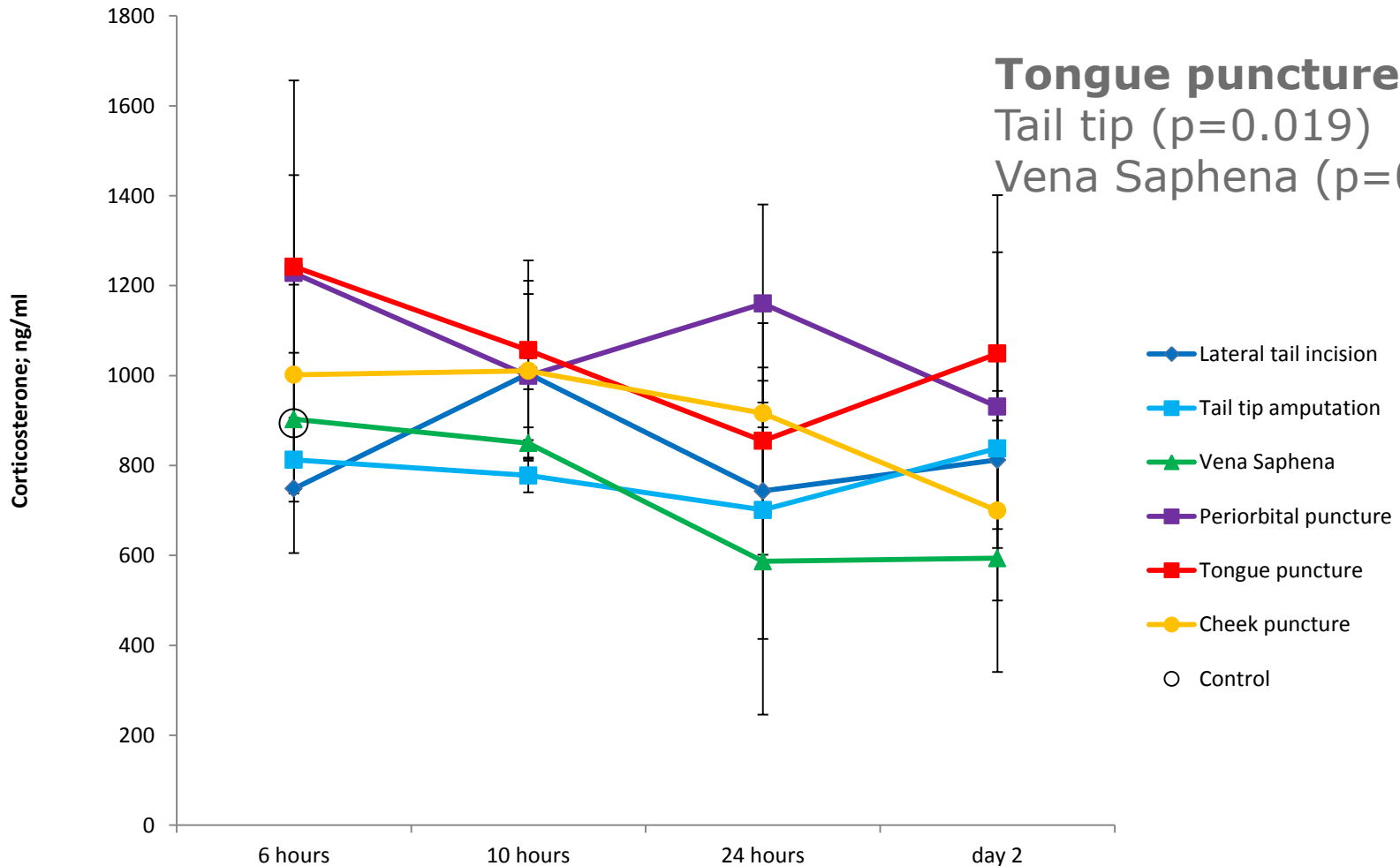
Stress hormone

Corticosterone (plasma)



Stress hormone

Corticosterone (plasma)



Periorbital puncture >

Tail inc. ($p=0.005$)

Tail tip ($p=0.001$)

Vena Saphena ($p<0.001$)

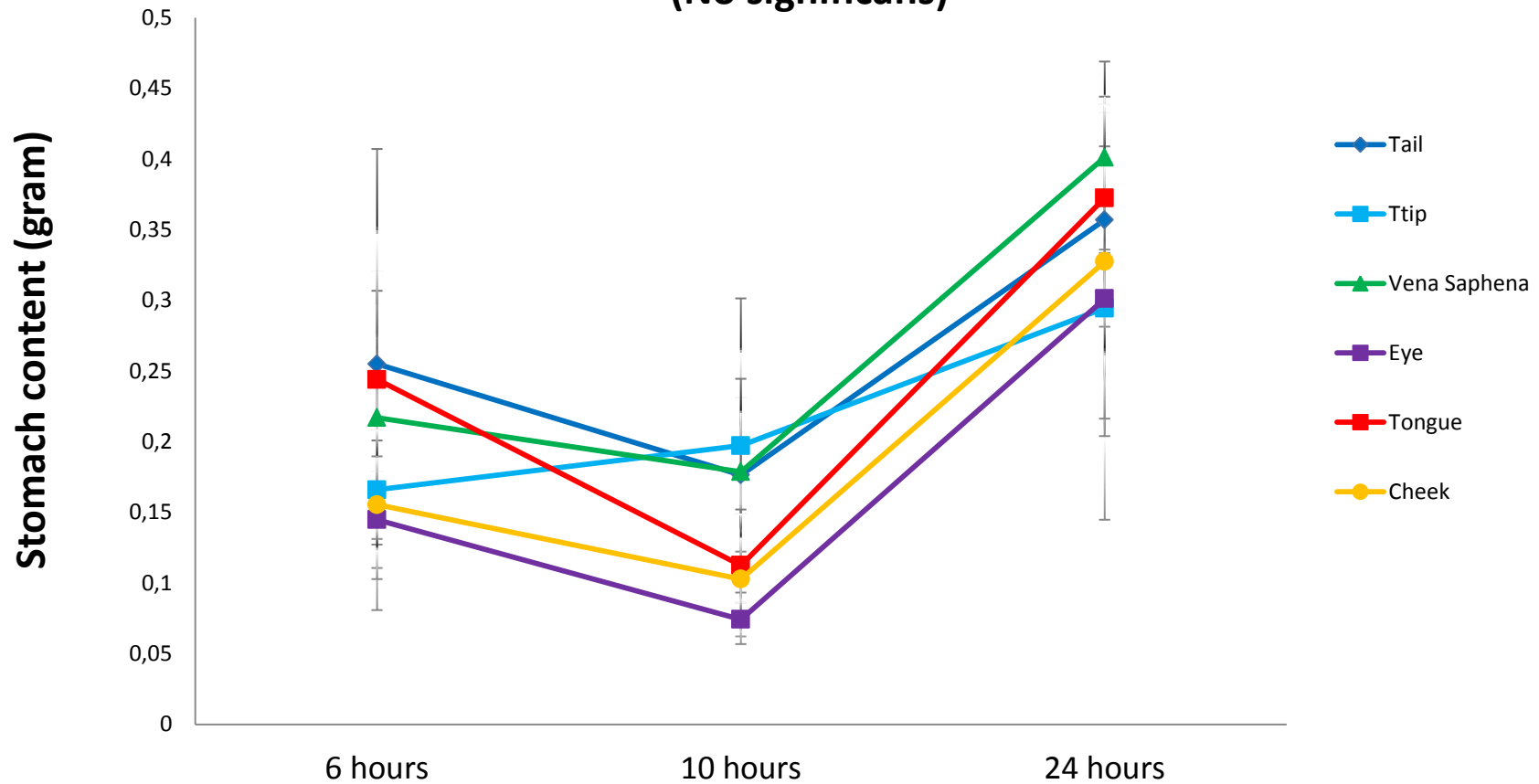
Tongue puncture >

Tail tip ($p=0.019$)

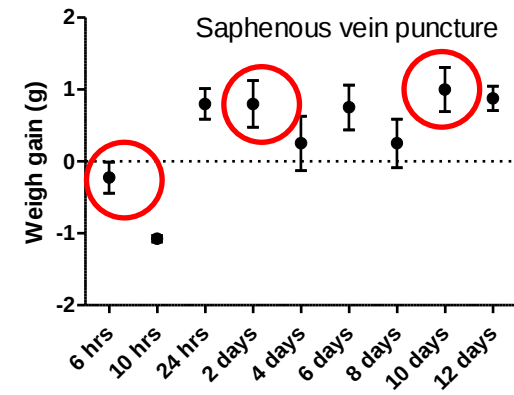
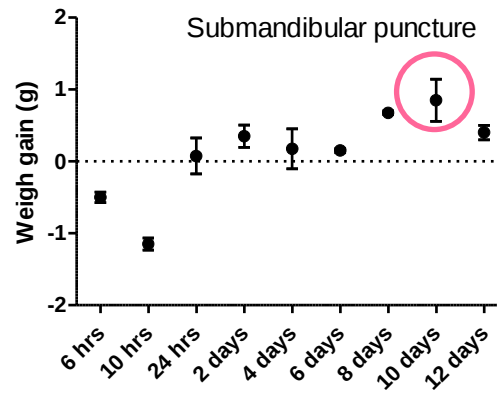
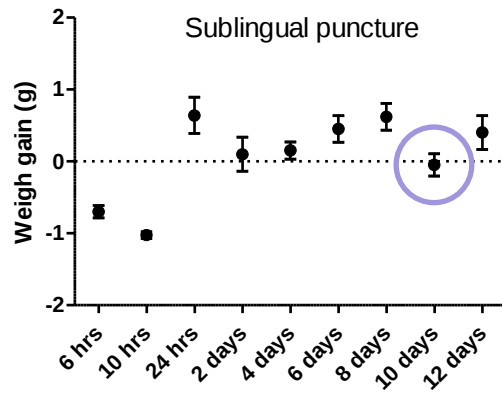
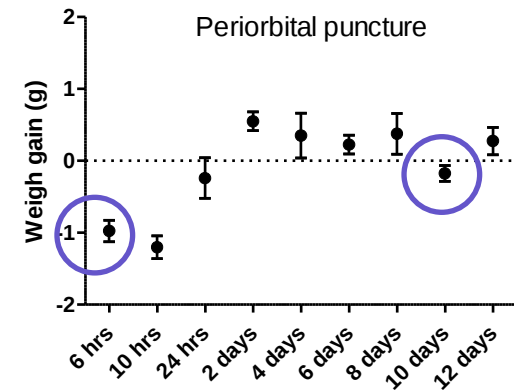
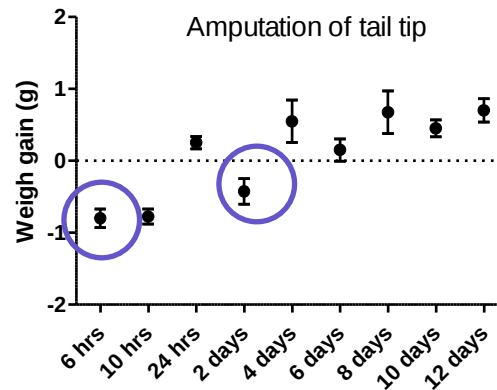
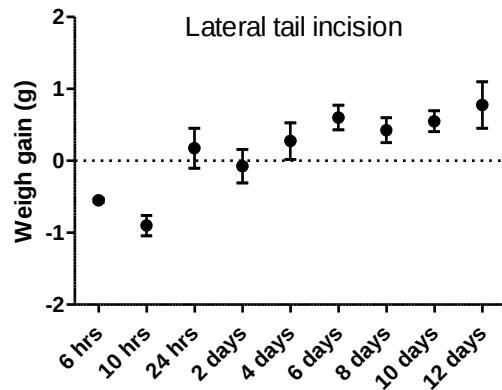
Vena Saphena ($p=0.003$)

Appetite/Eating behaviour

Stomach content after bloodsampling (No significans)



Body weight



6 methods – 3 assessment points

Histopathology

Lesions,
inflammation and
healing

Histopathology

Healing/inflamm: Avoid **Tailtip**
Abscesses, granulomas,
necrosis: Avoid **vena
sublingualis (Tongue)**

Markers of Inflammation

Expression of
inflammatory genes

Markers of Inflammation

Go for **vena Sublingualis**
(Tongue)
Avoid **Caudal region**

Animal Welfare

Stress, appetite and
body weight

Animal Welfare

Go for **vena Saphena**
Stress: Avoid **Eye + Tongue**
Appetite: Avoid **Head region**
Body w: Avoid **Eye + Tailtip**



The best way of blood sampling mice

Research Question:

Which method is the best when taking a blood sample in mice?

Short answer:

Long answer:



The best way of blood sampling mice

Research Question:

Which method is the best when taking a blood sample in mice?

Short answer:

It depends...

Long answer:



The best way of blood sampling mice

Research Question:

Which method is the best when taking a blood sample in mice?

Short answer:

It depends...

Long answer:

It's complicated...



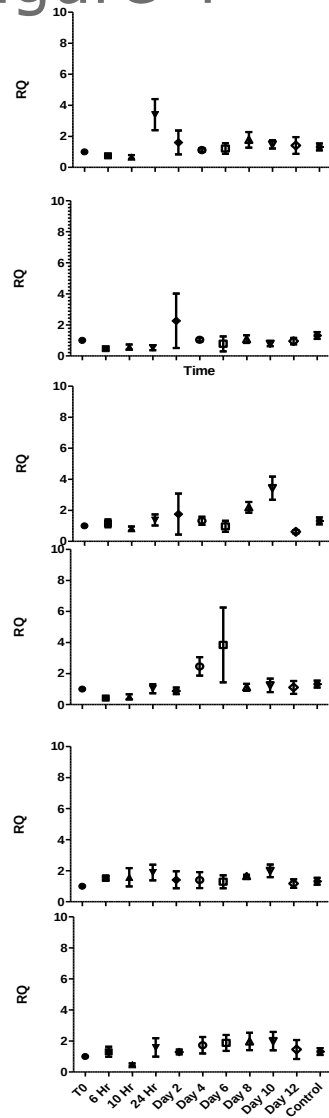
Acknowledgement

Animal technicians Mette Nelander, Anni Mehlsen and Helene Farlov

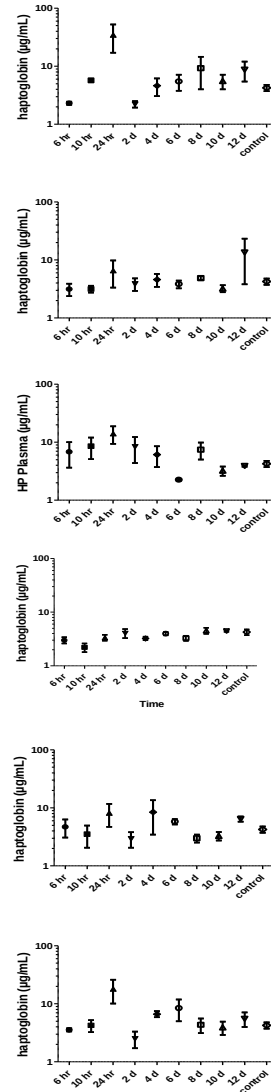


Figure 4

A



B

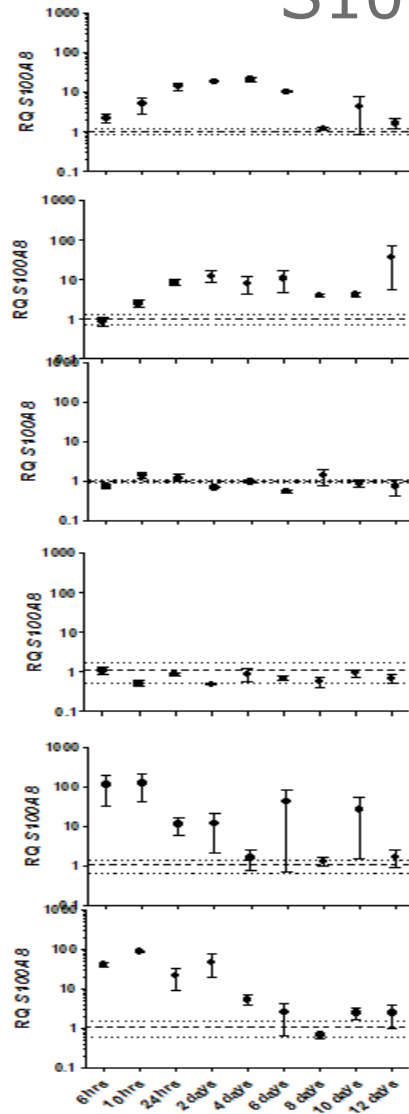


Haptoglobin;
plasma

Figure 9

A

S100A8



B

Cxcl2

Lateral tail incision

Tail tip amputation

Periorbital puncture

Sublingual puncture

Cheek puncture

V. saphena puncture

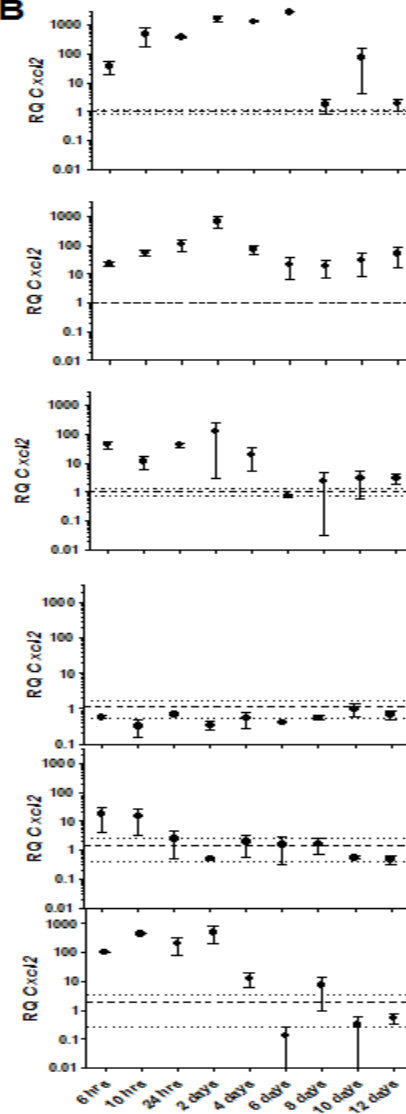
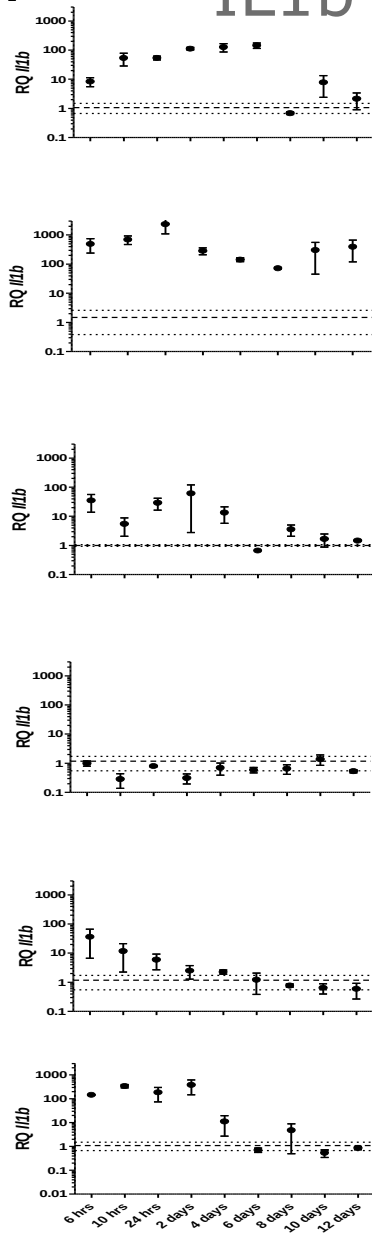


Figure 10

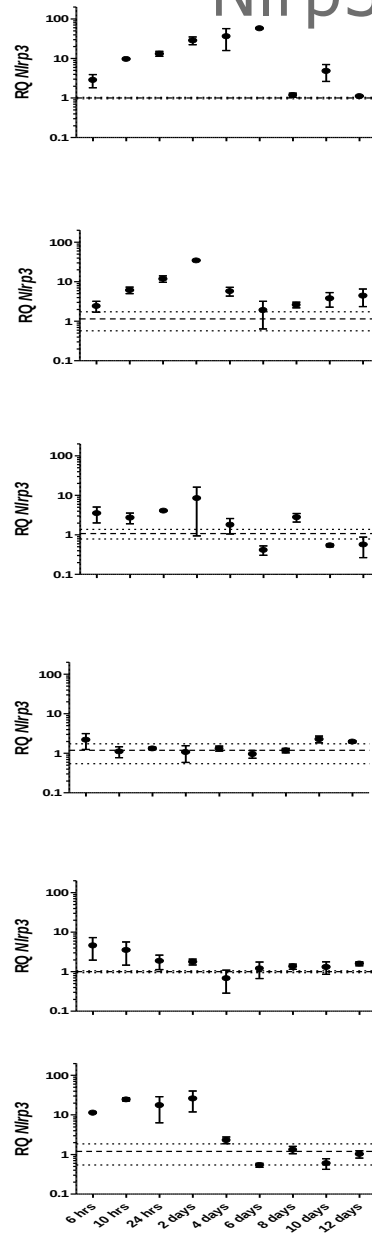
A

IL1b



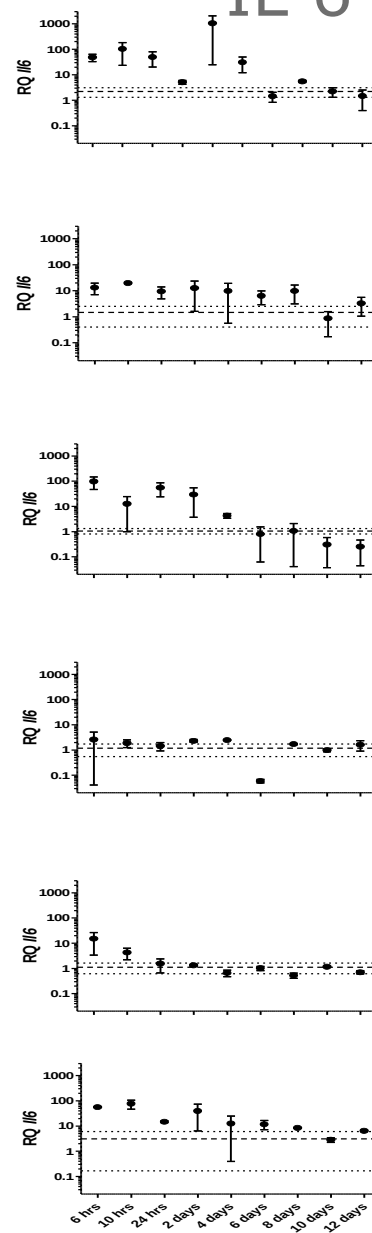
B

Nlrp3



C

IL-6



Lateral tail incision

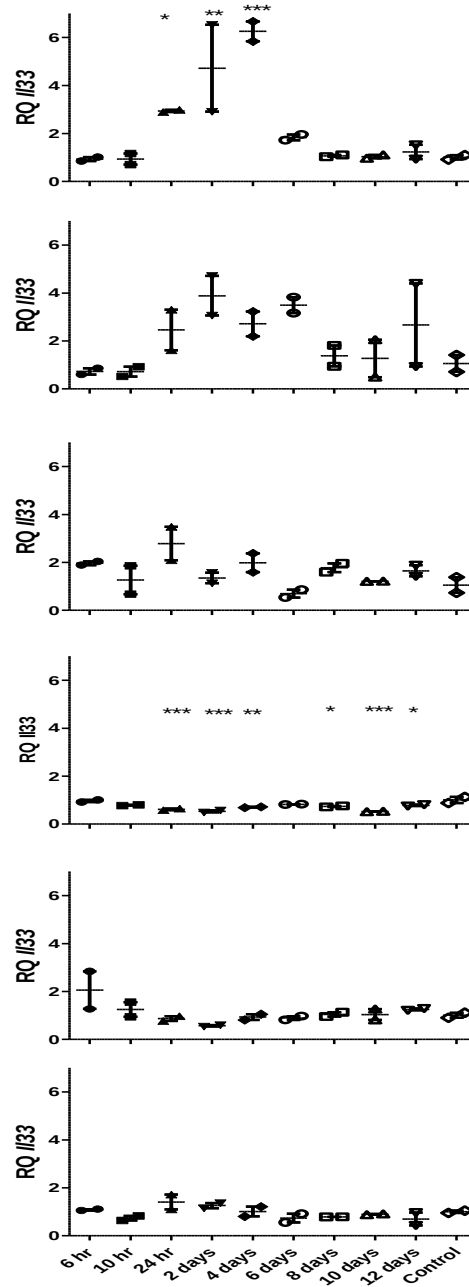
Tail tip amputation

Periorbital puncture

Sublingual puncture

Cheek puncture

Figure 11 IL-33



Lateral tail incision

Tail tip amputation

Periorbital puncture

Sublingual puncture

Cheek puncture

Figure 1

Figure: Weight gain

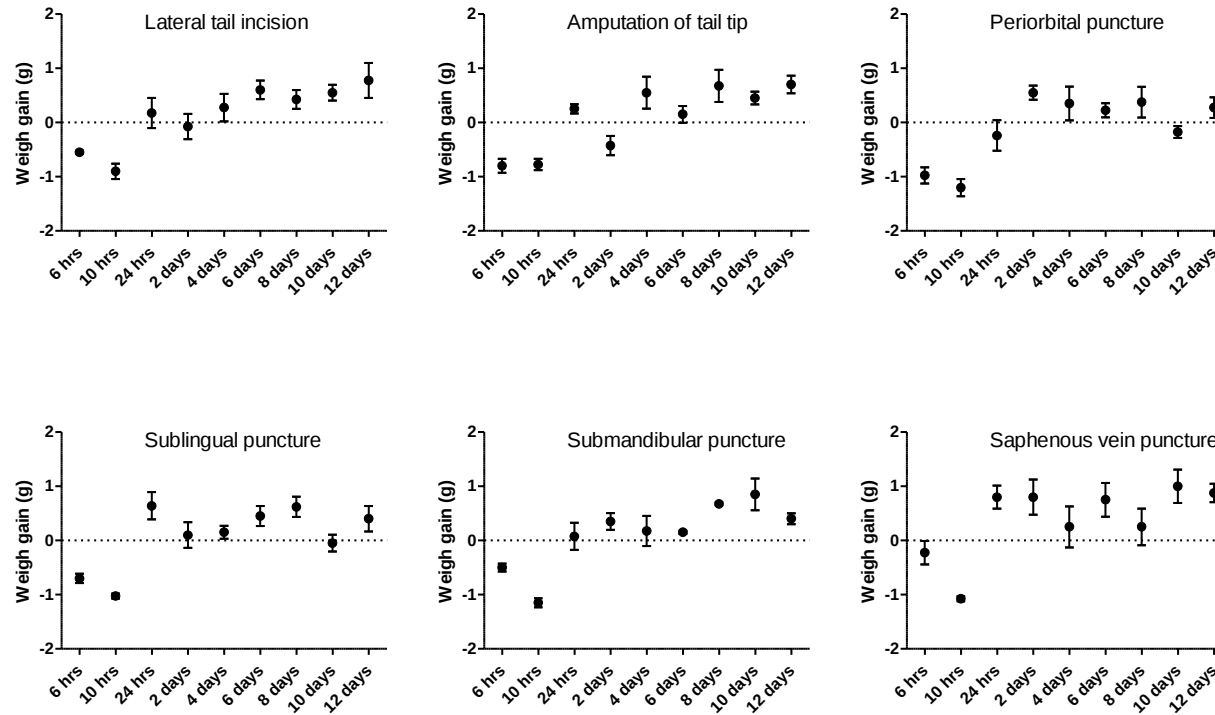


Figure 2

Figure: Stomach content

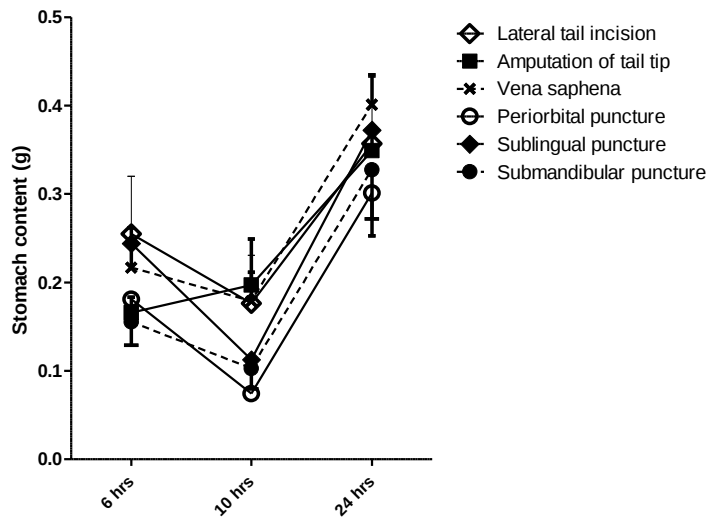


Figure: Weight gain/Stomach content correlation

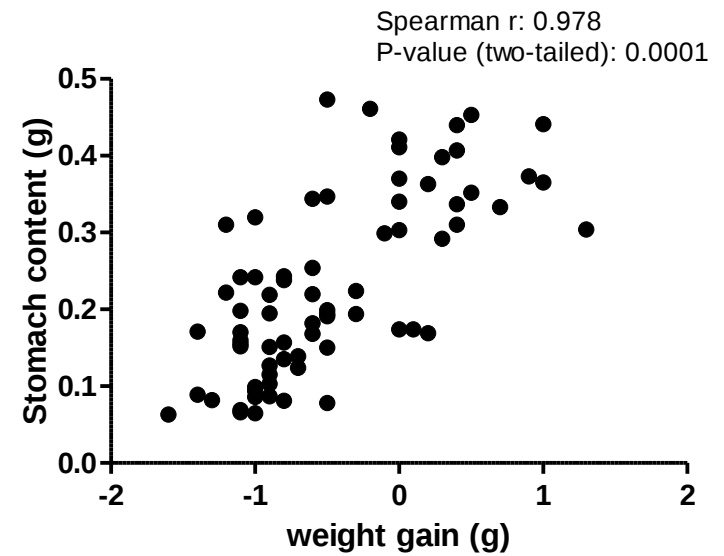


Figure 3

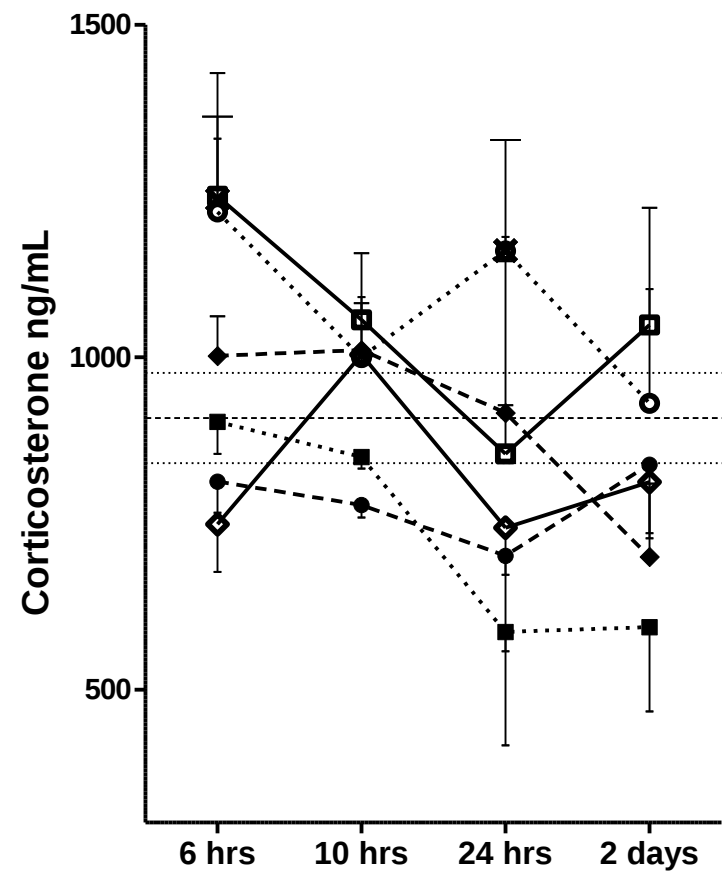
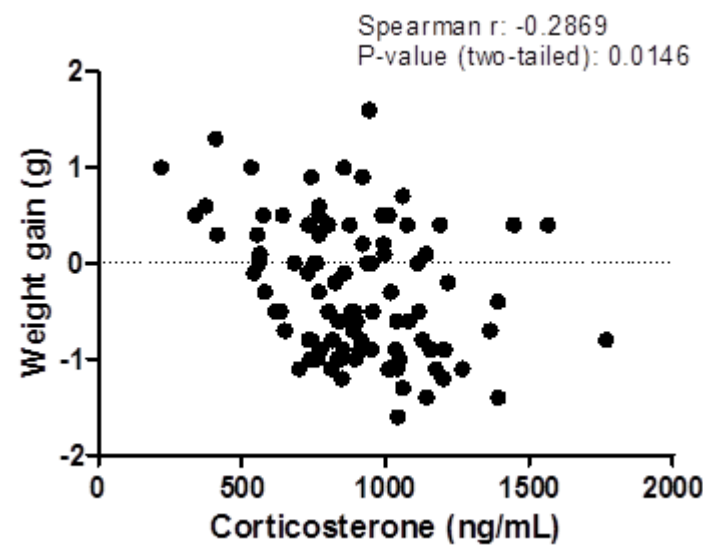
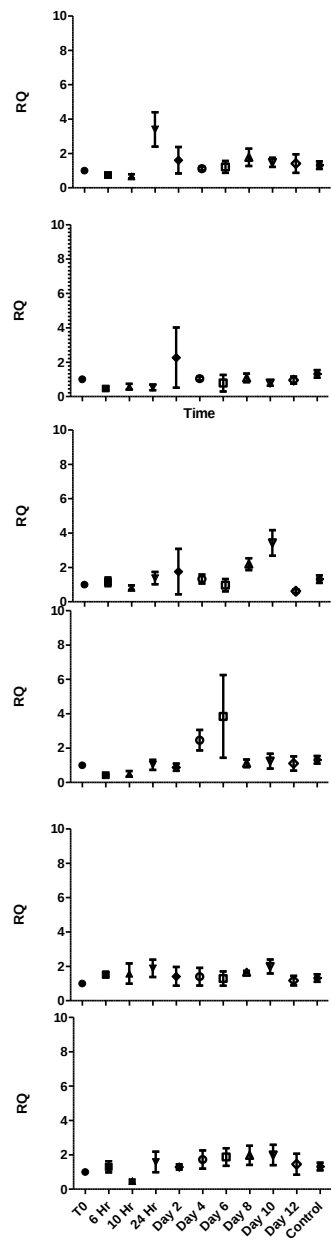


Figure: Corticosterone/Weight gain correlation



A



B

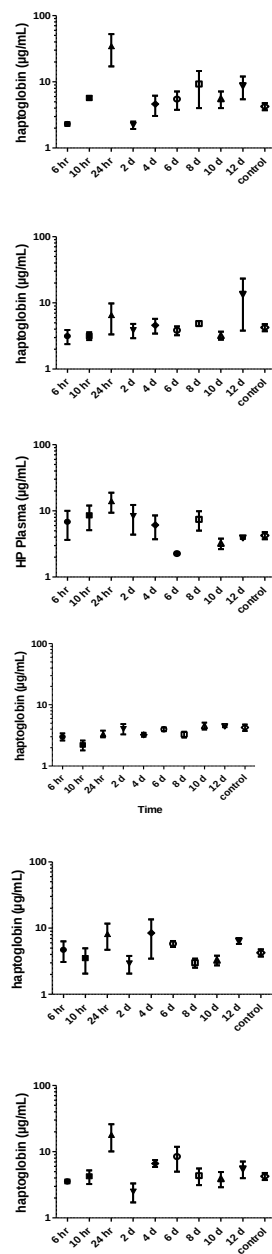
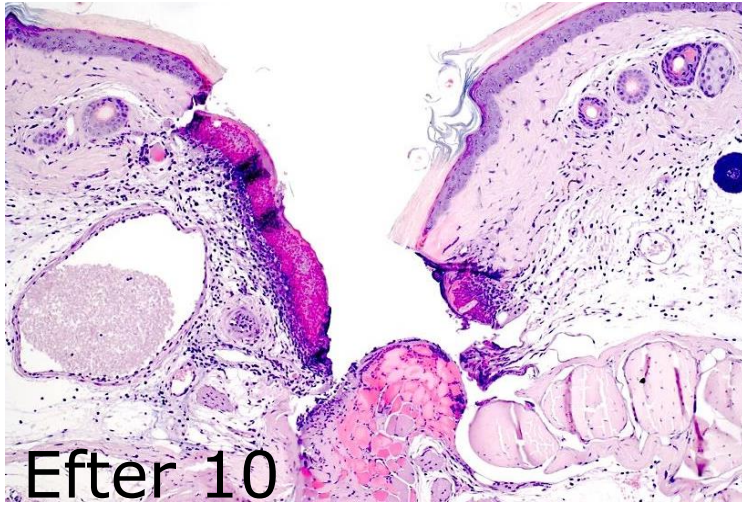
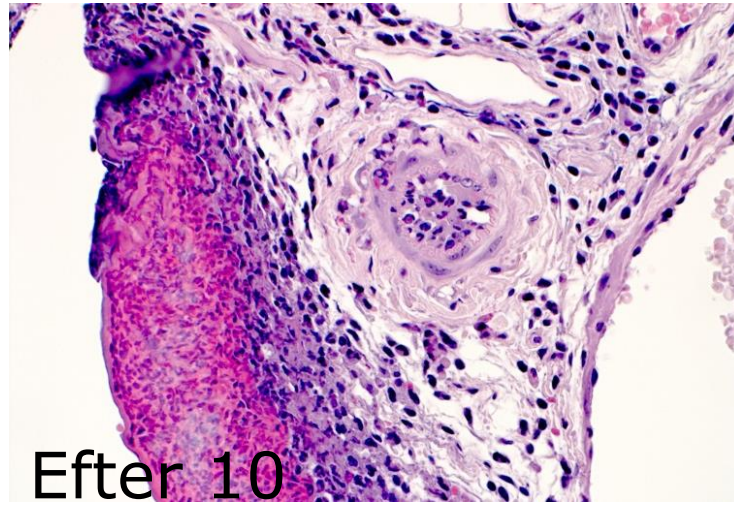


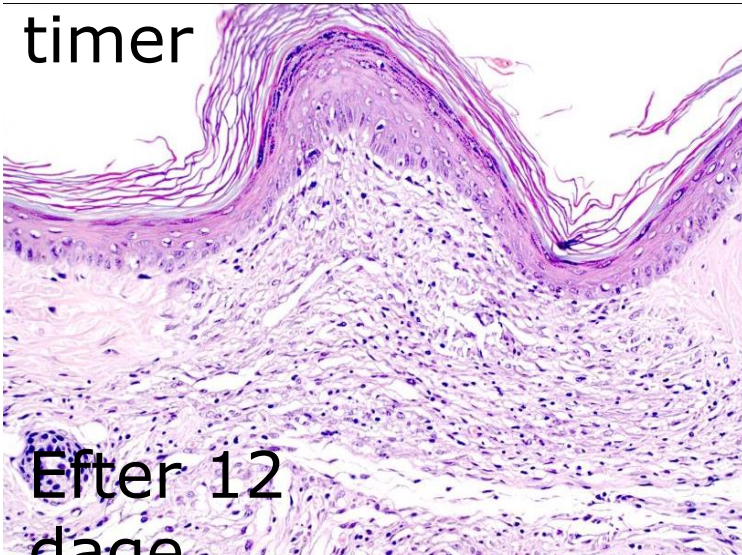
Fig 5 **Lateral halesnit**



Efter 10
timer

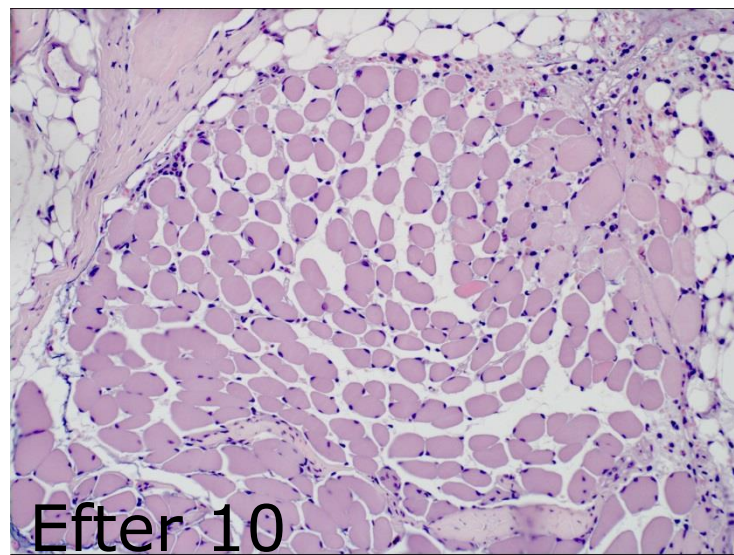


Efter 10
timer



Efter 12
dage

Sted og dato



Efter 10
timer

Fig 6 **Halespids**

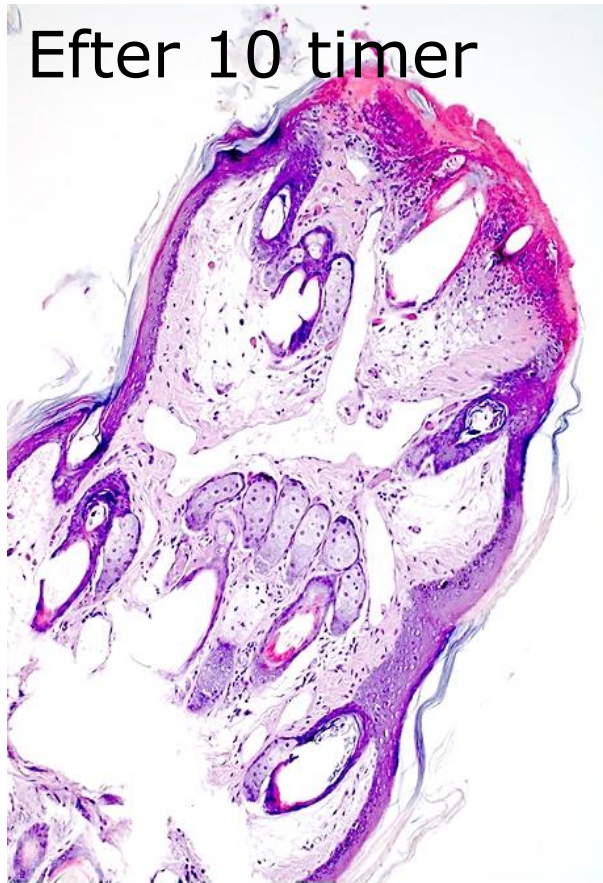
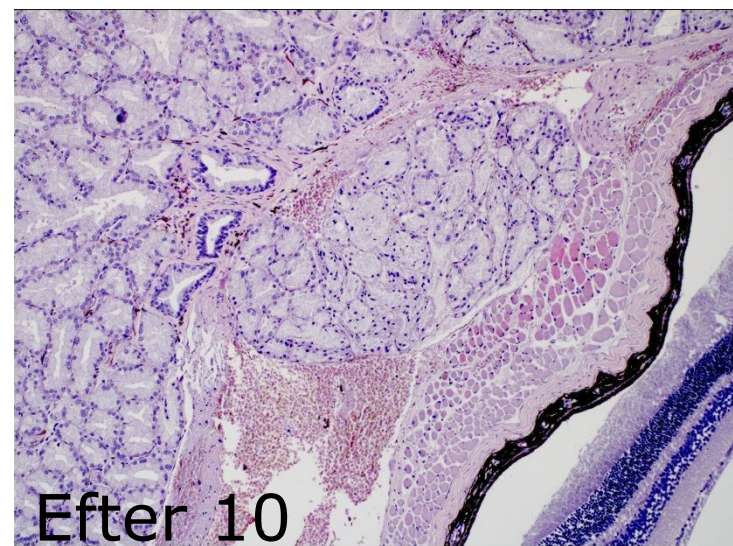
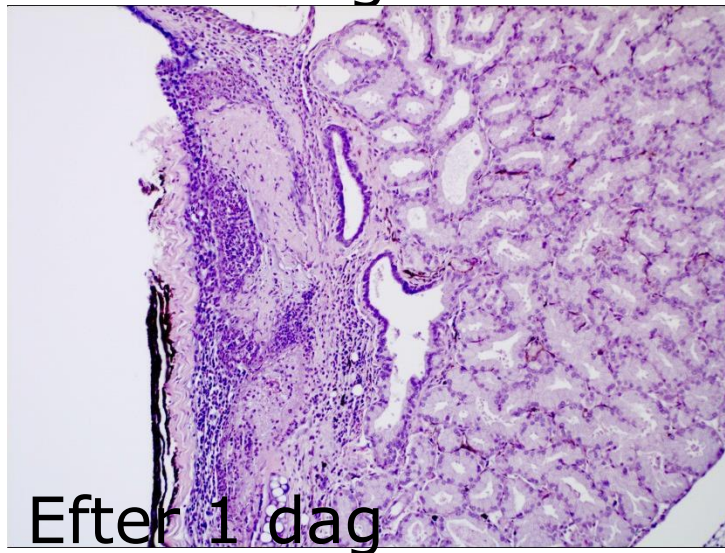
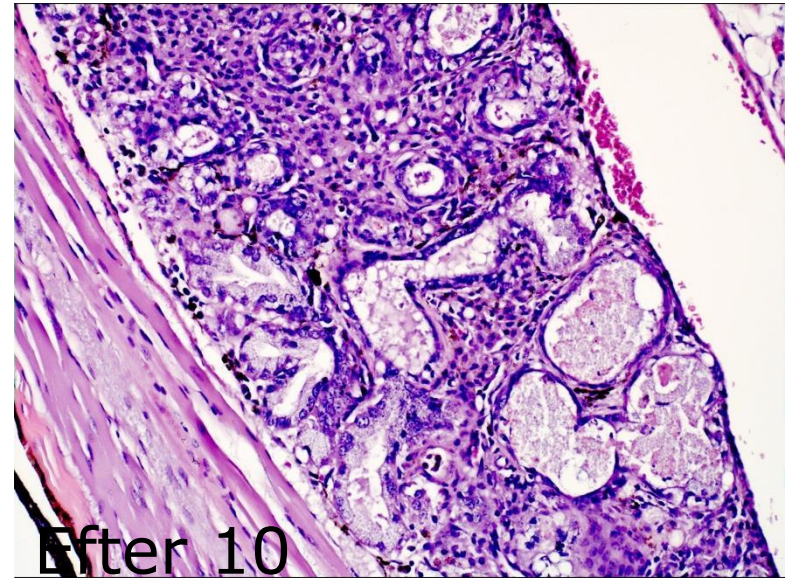
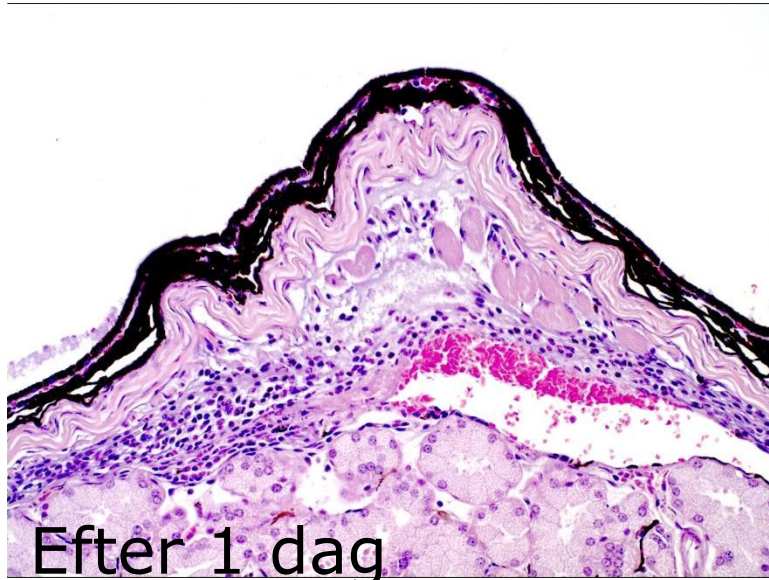


Fig 7
Øje

timor

Fig 8 Tunge

