Selection of rainbow trout (*Oncorhynchus mykiss*) lines for divergent stress responsiveness

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The stress response is essentially adaptive - why modify?

To reduce behaviours/responses which are inappropriate, or are associated with welfare problems.

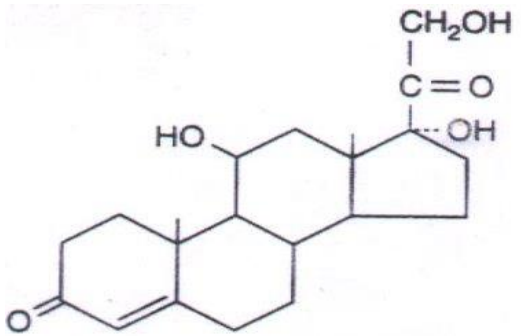
Stress is unavoidable under finfish aquaculture conditions.

Stress = ↓growth; ↓ reproduction; ↓ immunocompetence; ↓ flesh quality.

How can we modify the stress response?

Cortisol = common factor

Reduced cortisol = reduced problem?



Cortisol

Outcomes:

- *increased production*
- *improved reproductive performance*
- *reduced incidence of disease*
- *improved "well-being" of captive animals*
- *accelerate "domestication"*

Therefore – reduce circulating cortisol during stressful events

EU project: Selective Breeding for Stress Tolerance in Aquacultured Fish

Project asked the questions:

Is the magnitude of the stress response a heritable trait in rainbow trout?

Is being a “low responder” an advantage under aquaculture conditions?

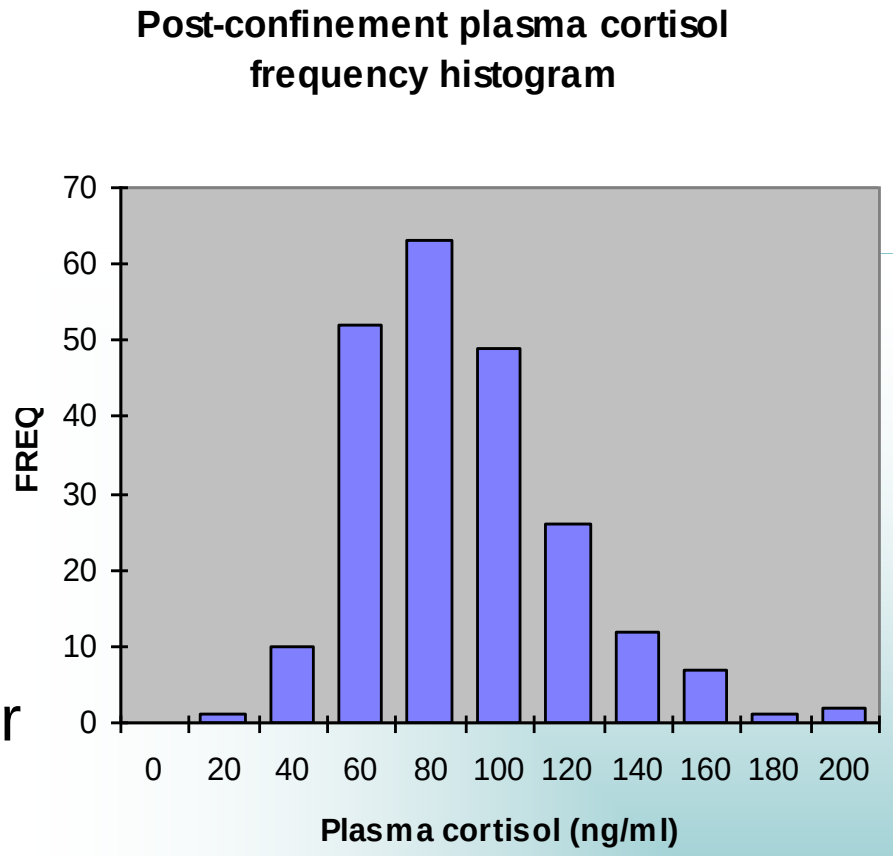
Are trout a suitable subject for selective breeding?

• Does stress responsiveness show broad variation within population?

Yes

• Is the level of stress responsiveness an individual characteristic that is stable over time?

Yes (for some of the population)



Do we know what intrinsic or extrinsic factors modify stress responsiveness?

Yes

- Environmental – *e.g. temperature*
- Social – *e.g. hierarchies*
- Developmental – *e.g. sexual maturity*

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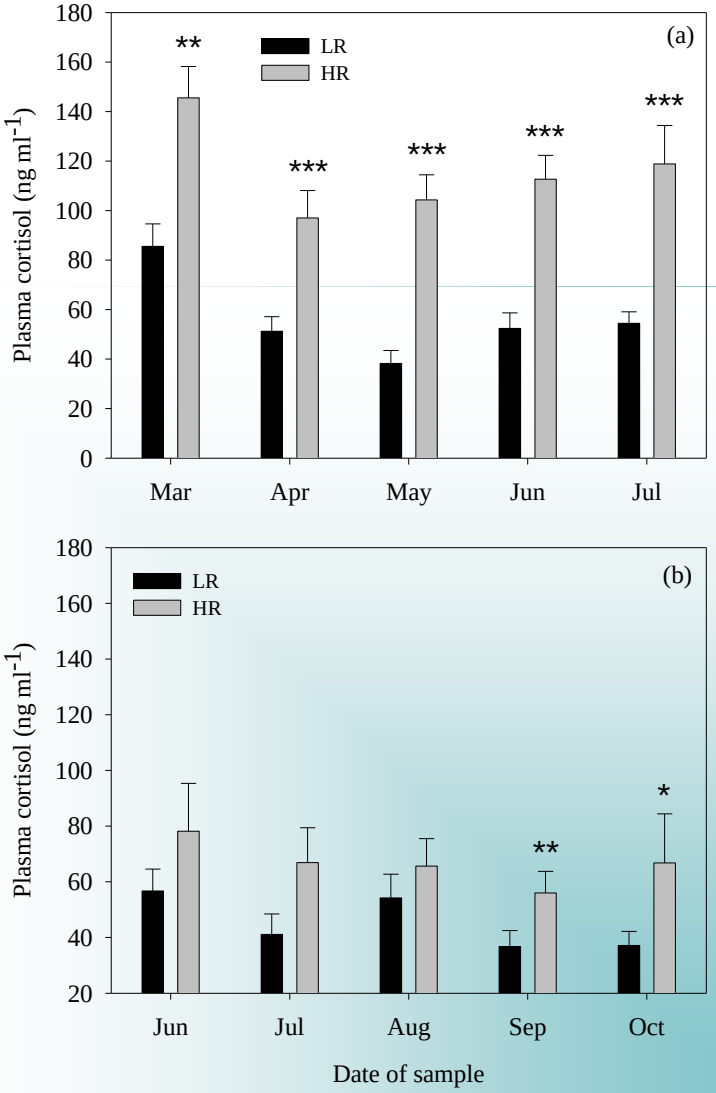
Establishing the lines:

- In 1996: 250 2+ rainbow trout PIT-tagged. Held as 25 fish/tank.
- Confined in small groups for 3 h at monthly intervals x 5
- Plasma cortisol levels determined.
- Mean plasma cortisol across all tests calculated for each fish.
- Fish ranked within each tank.
- Top 4 (HR) and bottom 4 (LR) fish in each tank selected.
- Progeny groups (families) generated from single male and female HR and LR parents (Feb 1997).
- Total of 14 LR and 15 HR families.

1996: F0 parental generation

Females

Males



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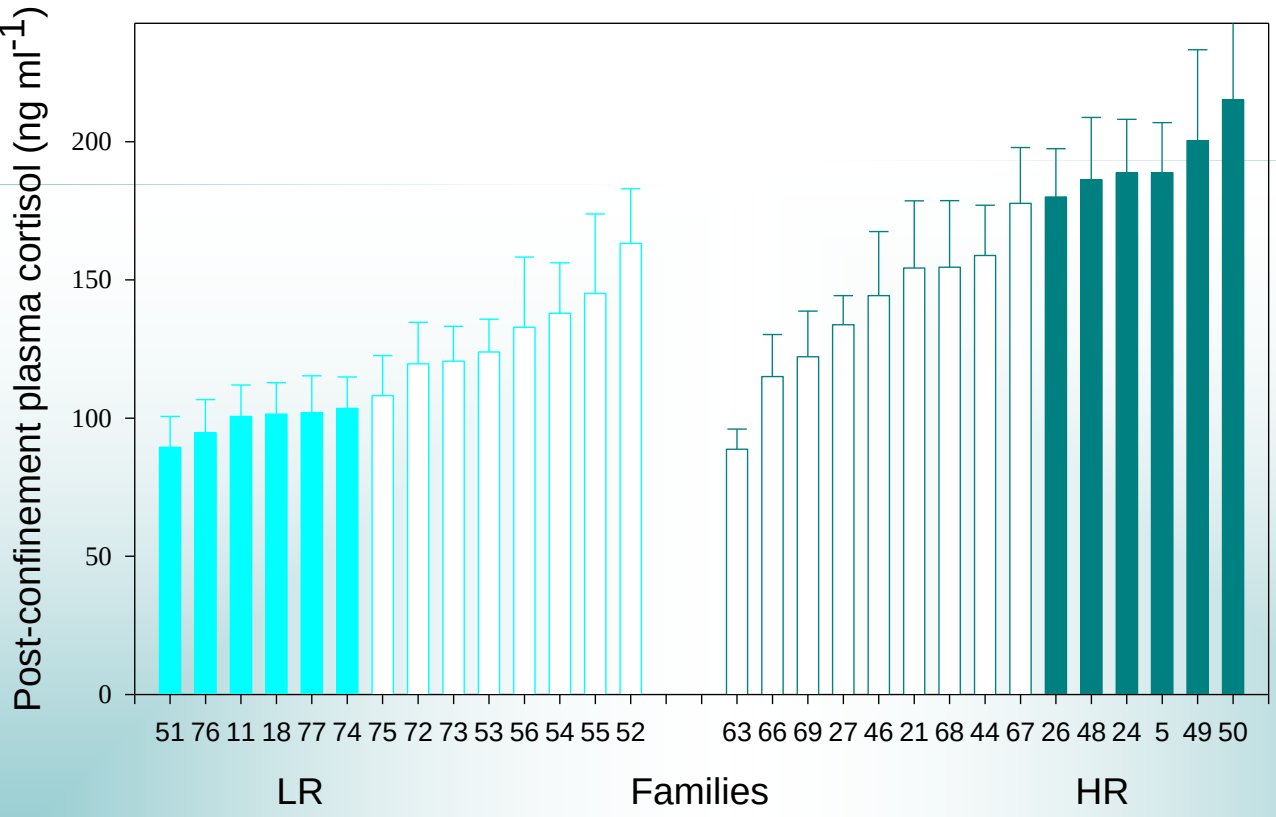
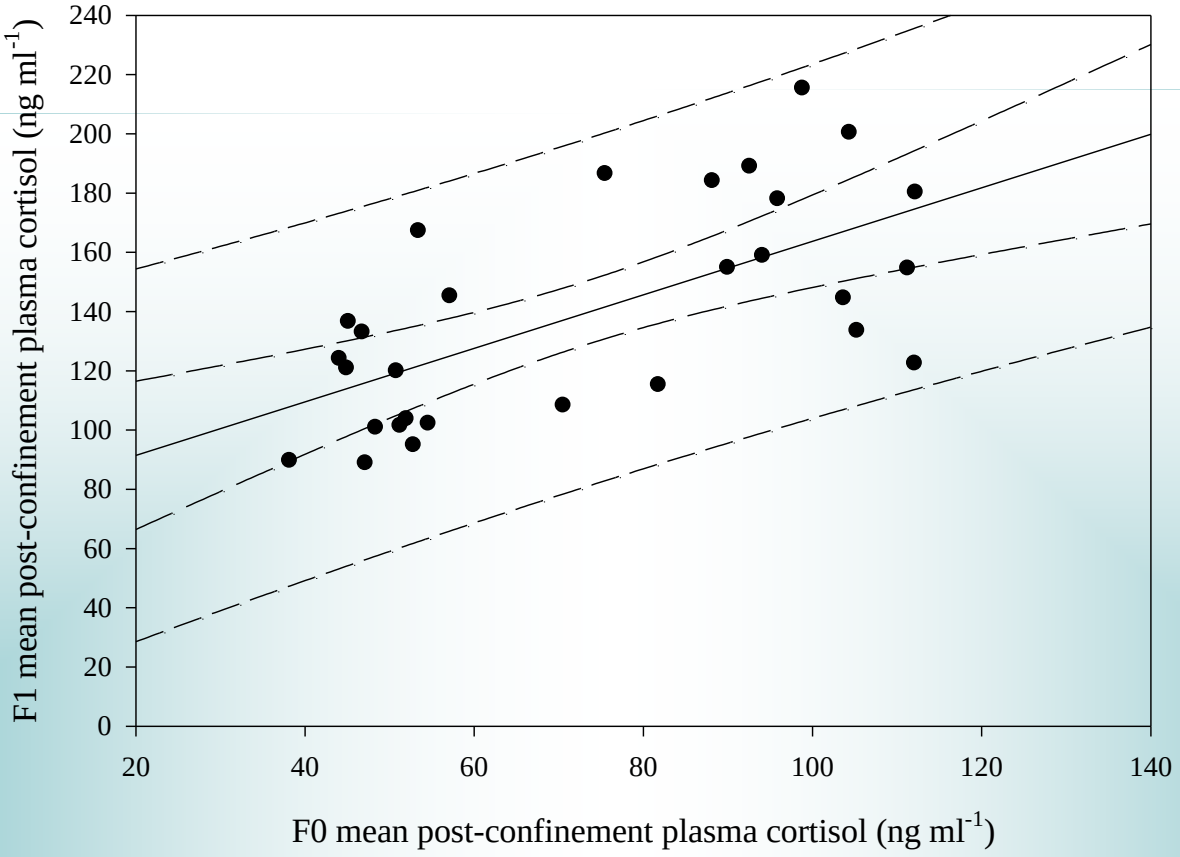
F1 (1997) progeny groups were tested by confinement on 5 occasions.

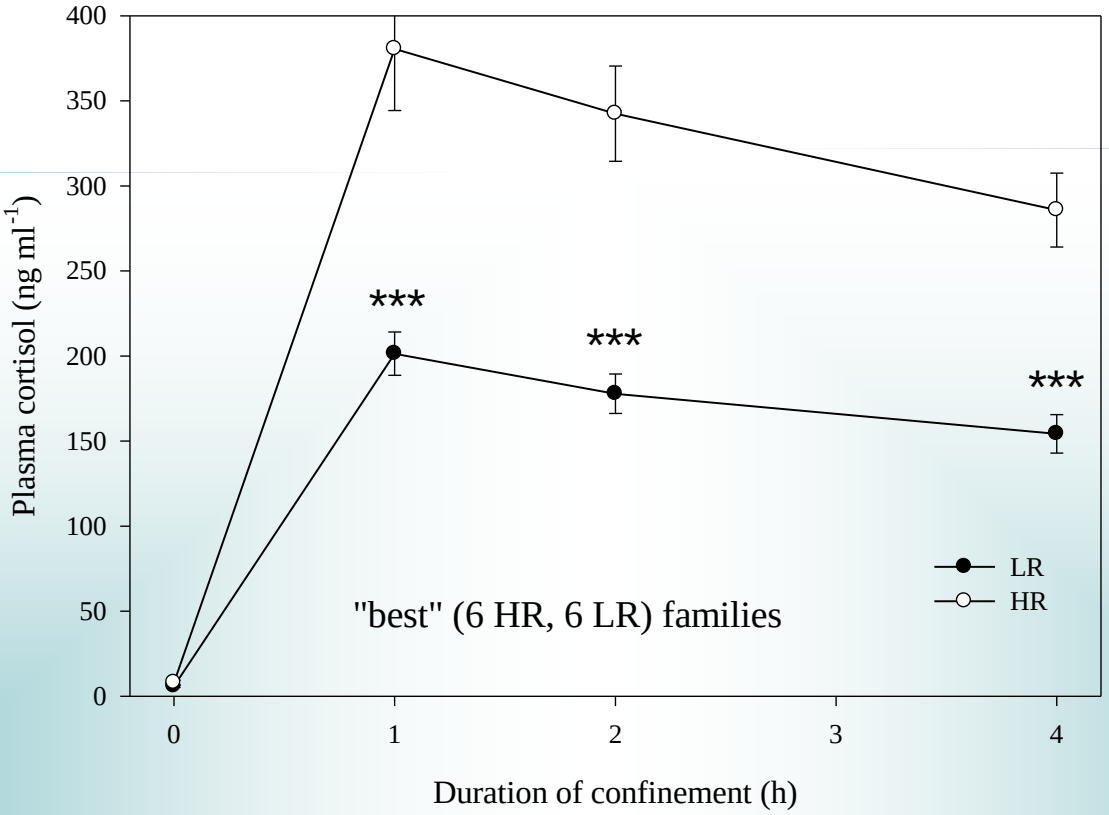


Heritability: Mean plasma cortisol response in each F1 progeny group plotted against mean F0 parent response ([male + female] / 2),

Estimated heritability $h^2 = 0.41$



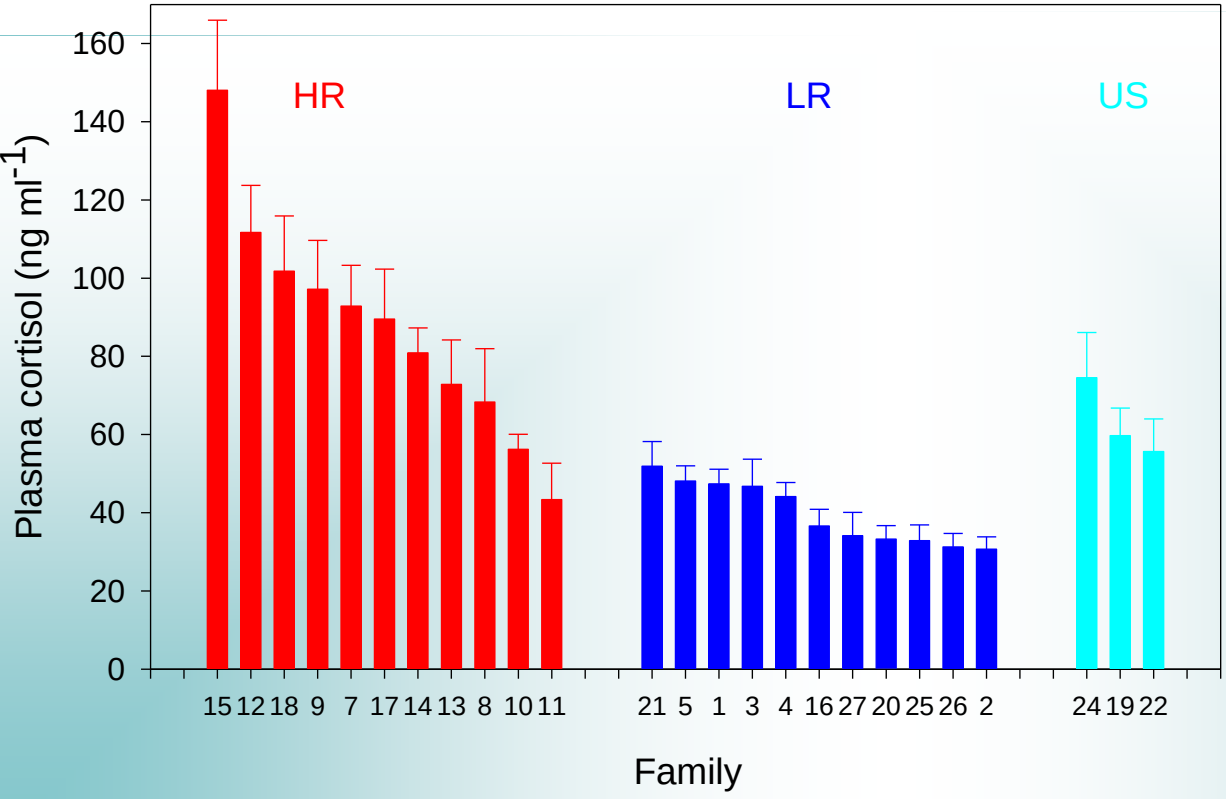
Lines exhibit divergent cortisol response to confinement.



F2 (2000) – Individual within family selection.

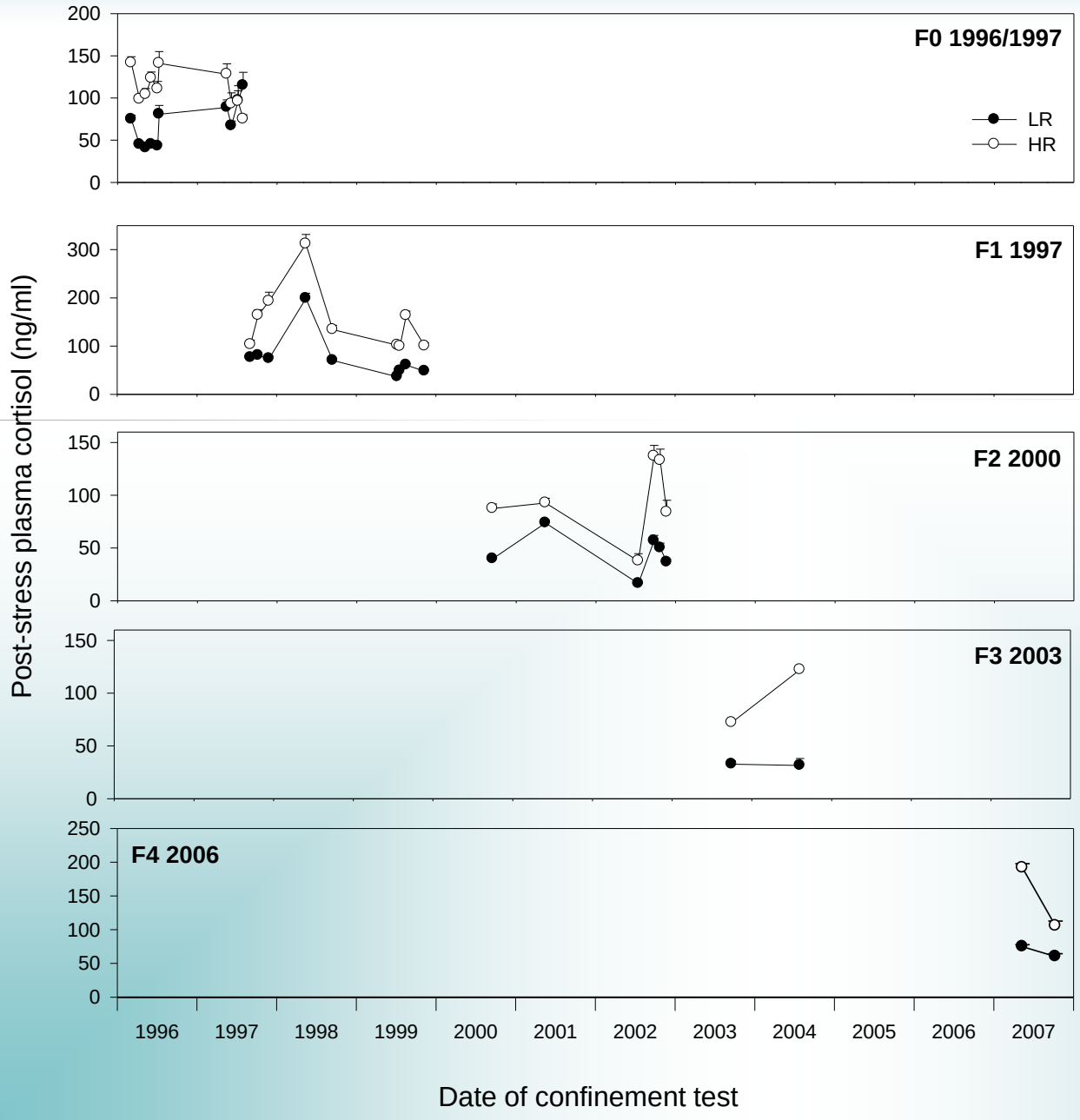
The two most divergent F1 families. Tested 3 times.
15 highest- and 15 lowest-responders selected.

Mean plasma cortisol levels following a 1h period of confinement:



F2 (2000) families

estimated h^2 = 0.6



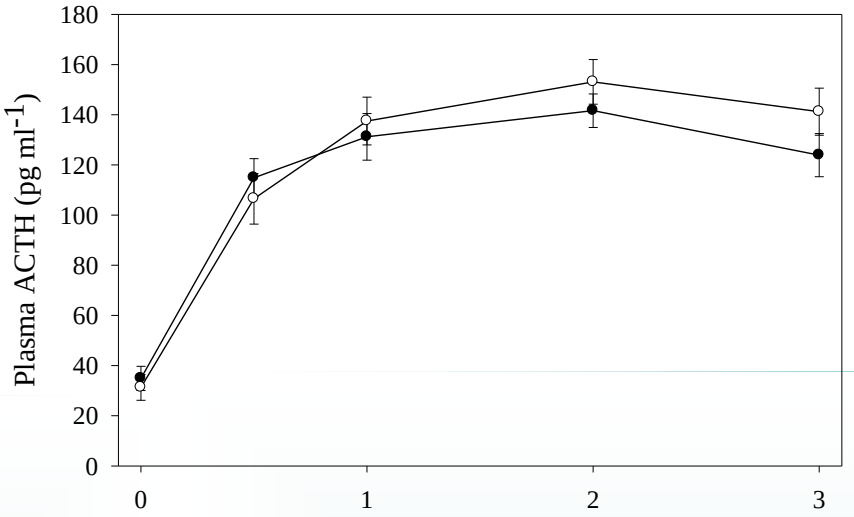
Stress response of HR & LR lines: Consistency with time

Significant variation within generations

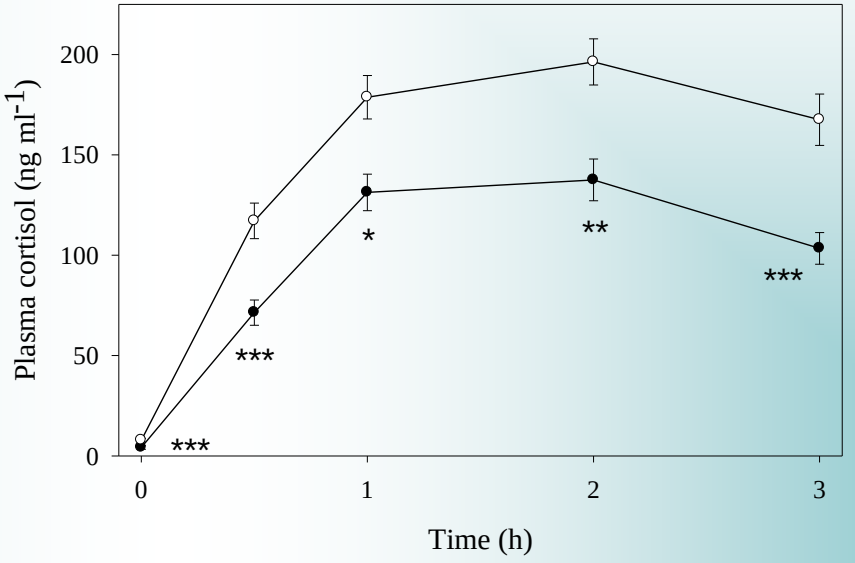
Degree of divergence not increased – limit reached?

Stress response of HR & LR lines:
Mechanistic basis

ACTH: HR = LR



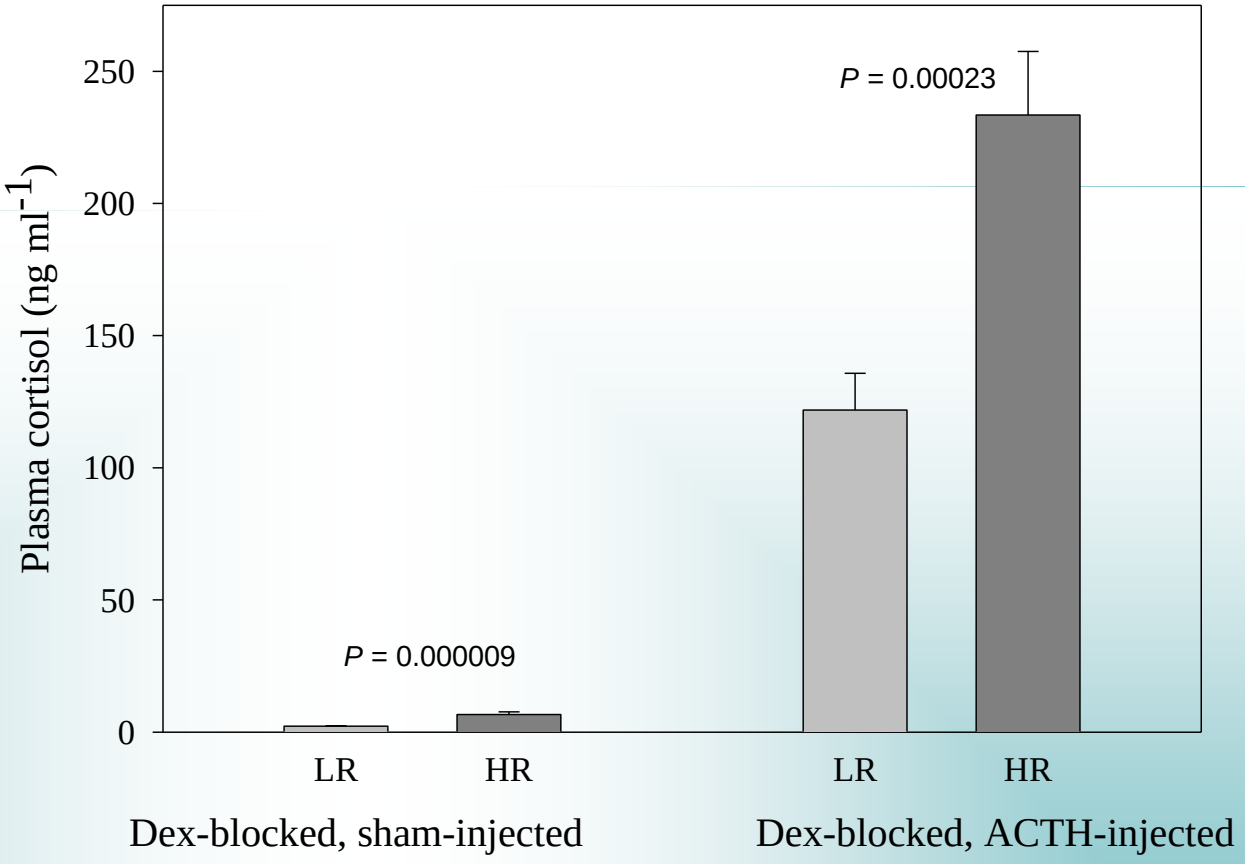
Cortisol: HR > LR



Interrenal function differs between lines?

Candidates:

$P450_{SCC}$?
StAR protein ?
ACTH receptor ?



Stress response of HR & LR lines *(F1, F2 & F3)* **:** Summary

- Plasma cortisol: **HR** > LR (F1-F4)
- Plasma epinephrine: **LR** > HR (F2)
- Plasma ACTH: **HR = LR !** (F1)

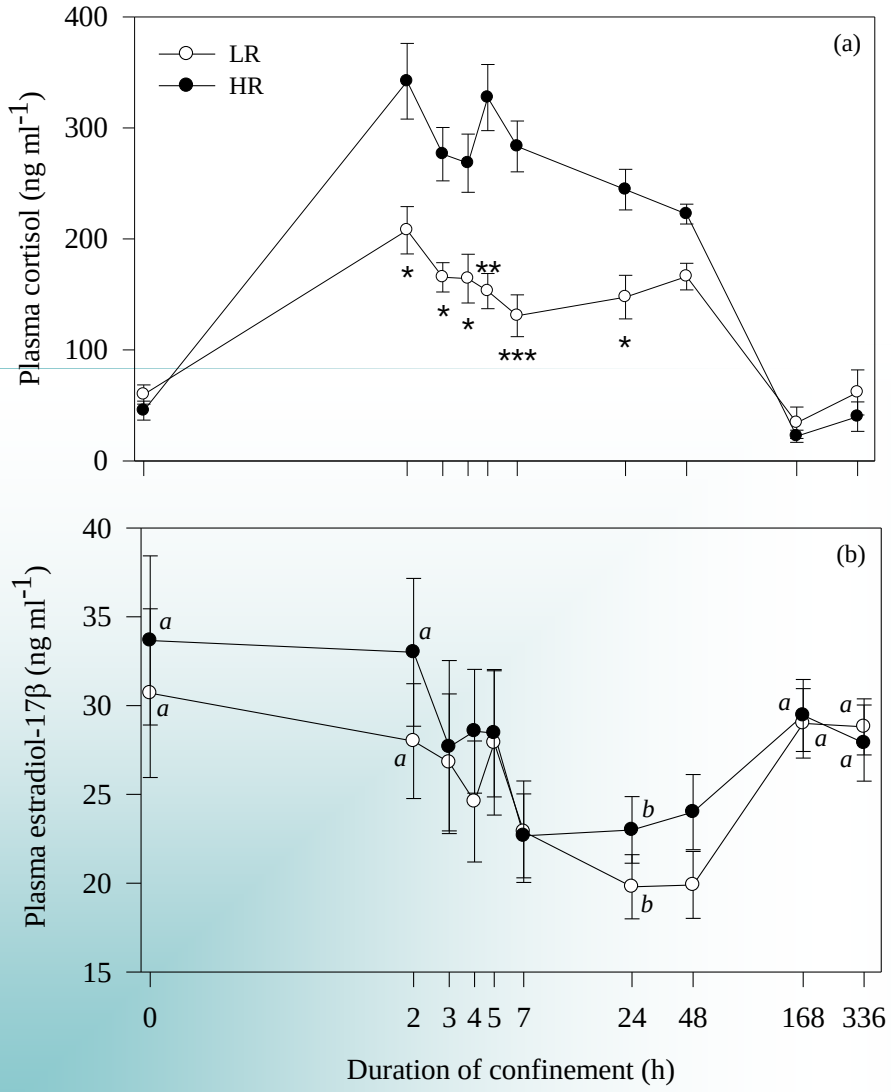
- Brain serotonergic activity: **LR** > HR (F2, F3)

- Plasma glucose: **LR** > HR (F2)
- Plasma lactate: **LR** > HR (F2)
- Plasma amino acids: **LR** > HR (F2)

- Plasma Na, K: HR = LR (F1)

- Hepatic cortisol binding: **recovery more rapid in LR (F2)**

Mature F0 females: changes in plasma cortisol and estradiol-17β during confinement

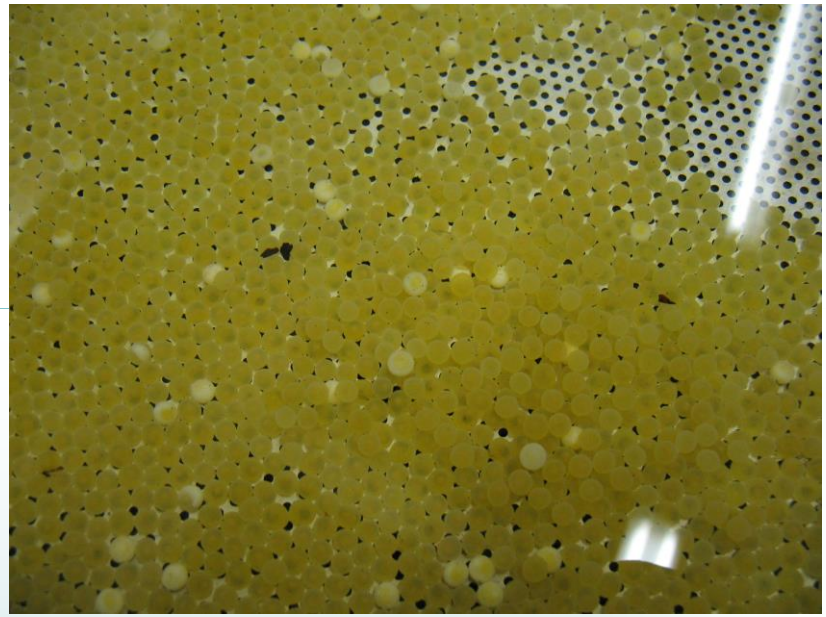


Reproductive performance of HR & LR lines (F1): Gonadal steroids

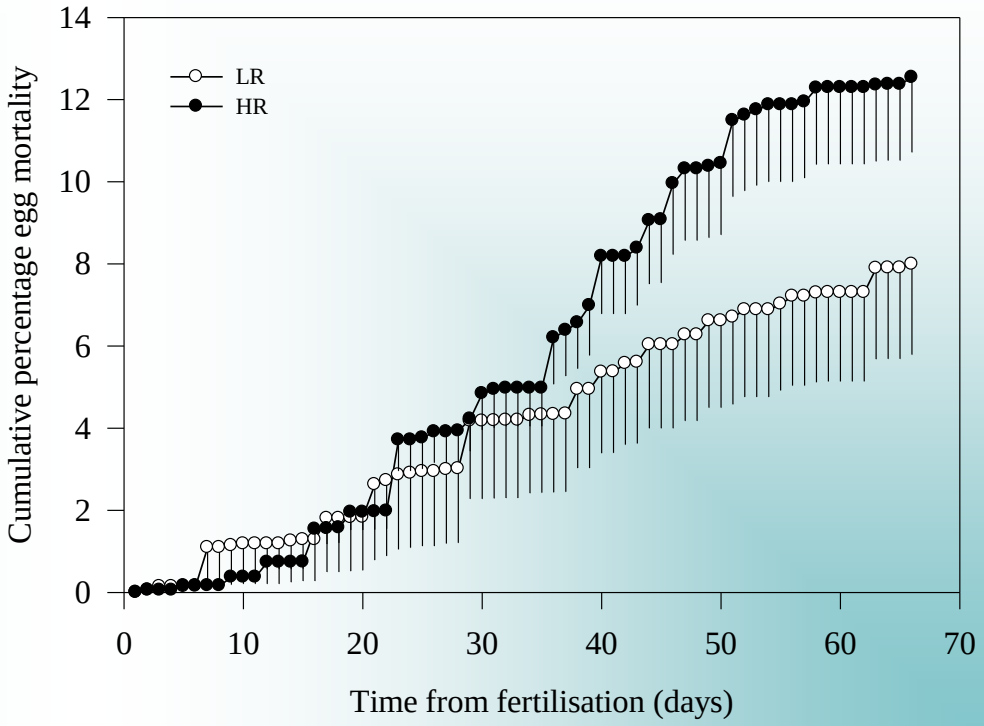
Cortisol: HR > LR

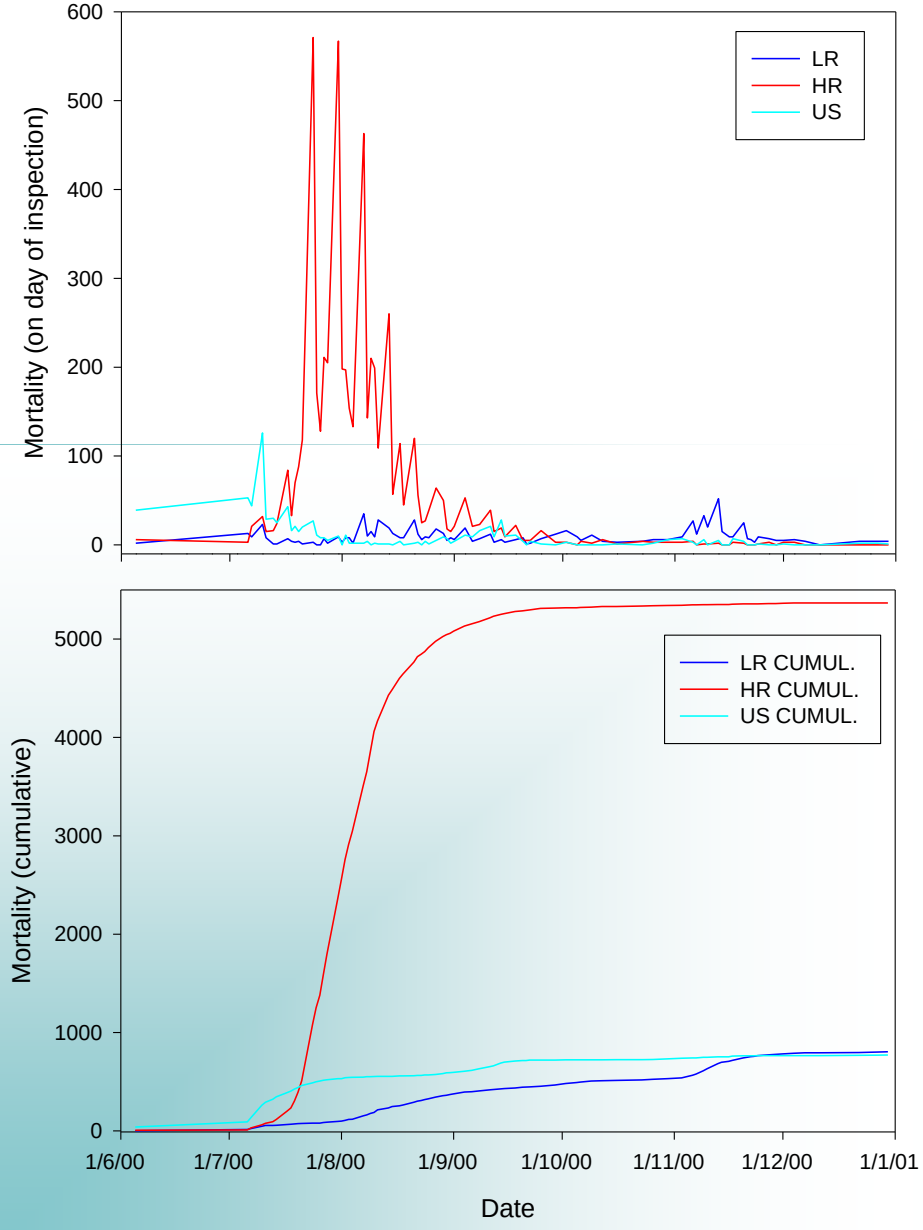
E2: HR = LR

Reproductive performance of HR & LR lines (F1): Gamete quality



Survival of fertilised ova (means of families)



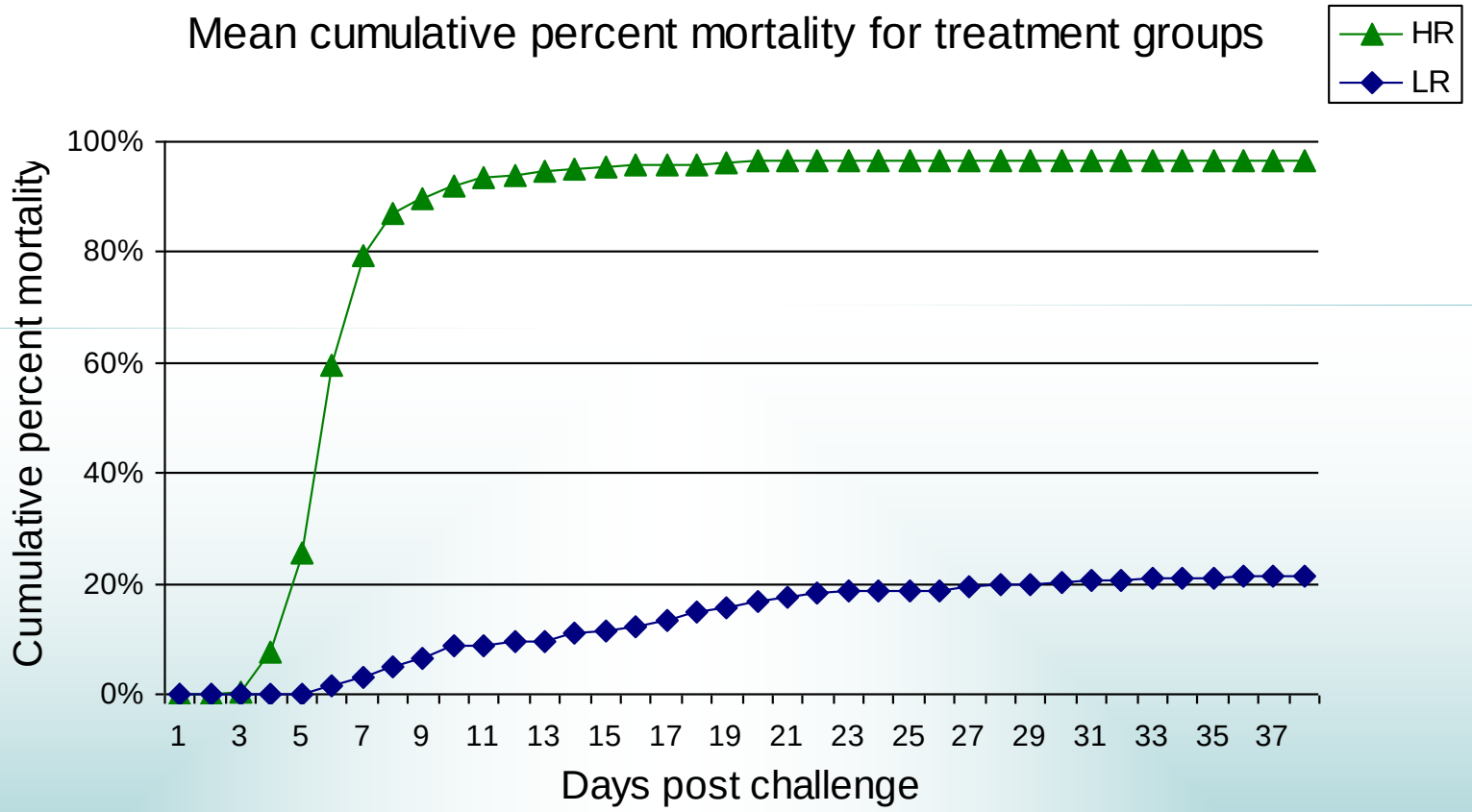


Reproductive performance of HR & LR lines (F1):
Fitness of progeny

Survival of progeny: LR > HR

True for all generations
Various causes

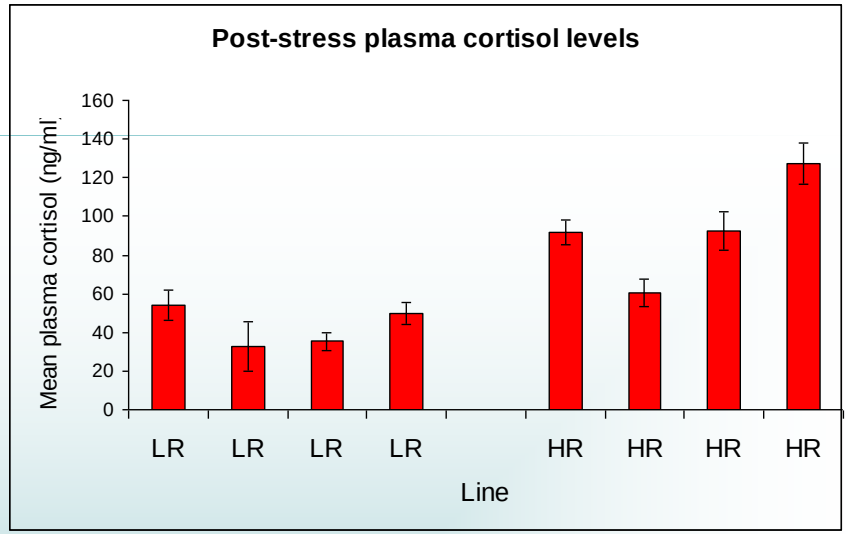
Disease resistance of HR & LR lines *(F2):*

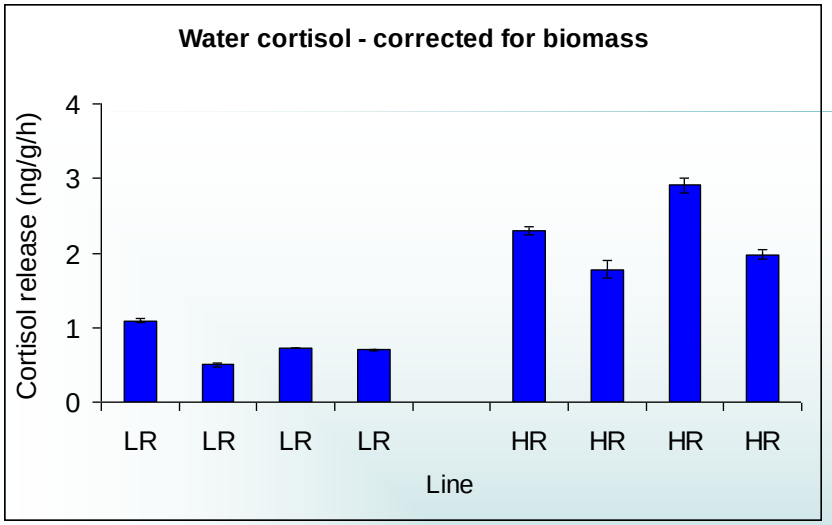


Reared from eggs at Cefas, Weymouth.
Four families of each line.
VHSV isolate freshwater strain 07-71 – bath challenge

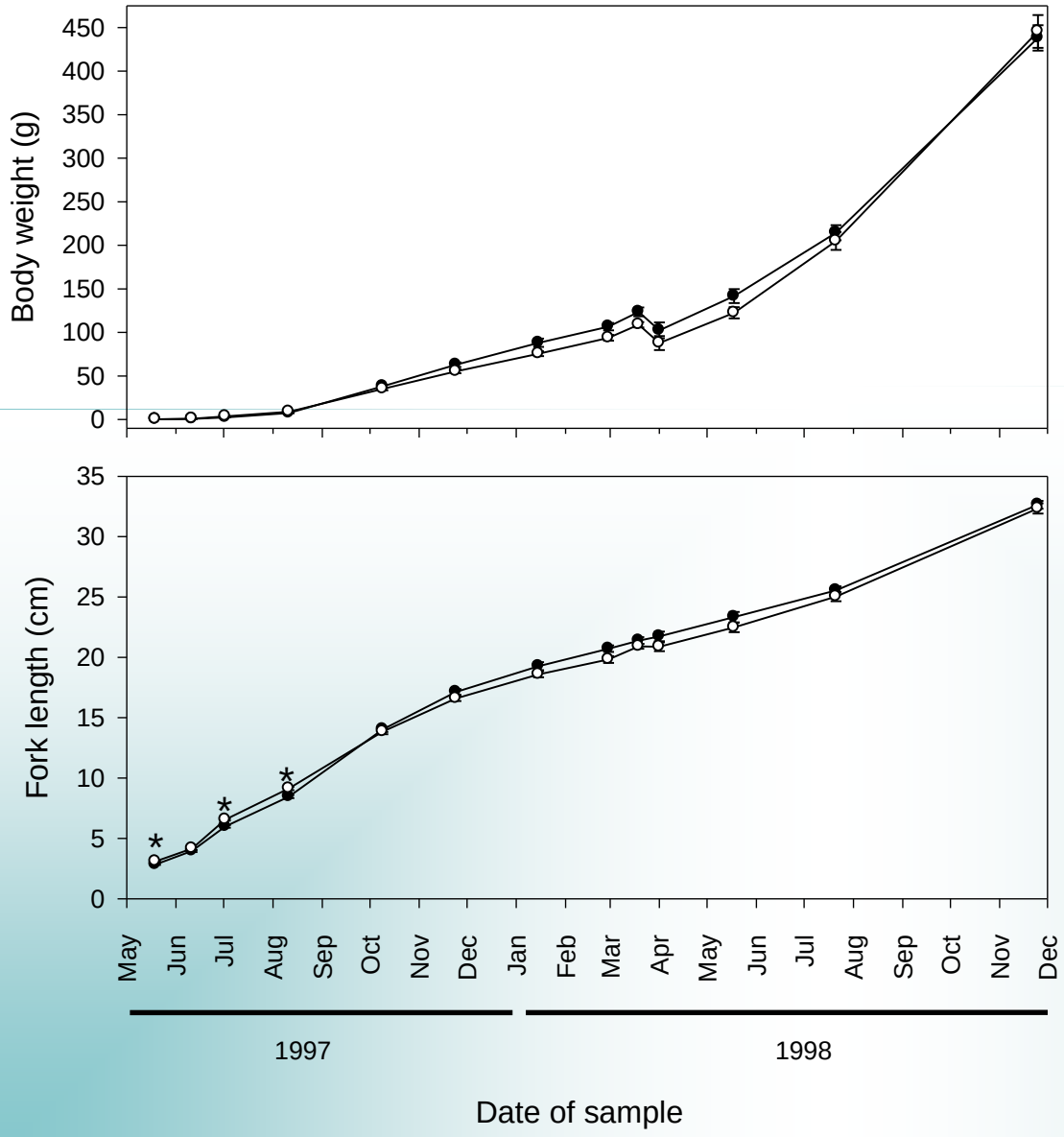
Disease resistance of HR & LR lines *(F2):*

Confirmation of divergence in stress responsiveness





Plasma cortisol = water-borne cortisol



Growth performance of HR & LR lines : *(F1)*

Reared as separate family groups

- ● LR
- ○ HR

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Reproductive and growth performance of HR & LR lines (F1): Summary

- Sperm count; Timing of ovulation; Fecundity: HR = LR
- Egg volume; Time to eyeing; Time to hatch: HR = LR
- ***Egg mortality: HR*** > *LR*
- ***Juvenile mortality: HR*** > *LR*
- Growth: HR = LR

Performance of HR & LR lines : *Conclusion*

Is the magnitude of the stress response a heritable trait in rainbow trout? ***Yes***

Is being a “low responder” an advantage under aquaculture conditions? ***Possibly – not a disadvantage (relative to HR)***

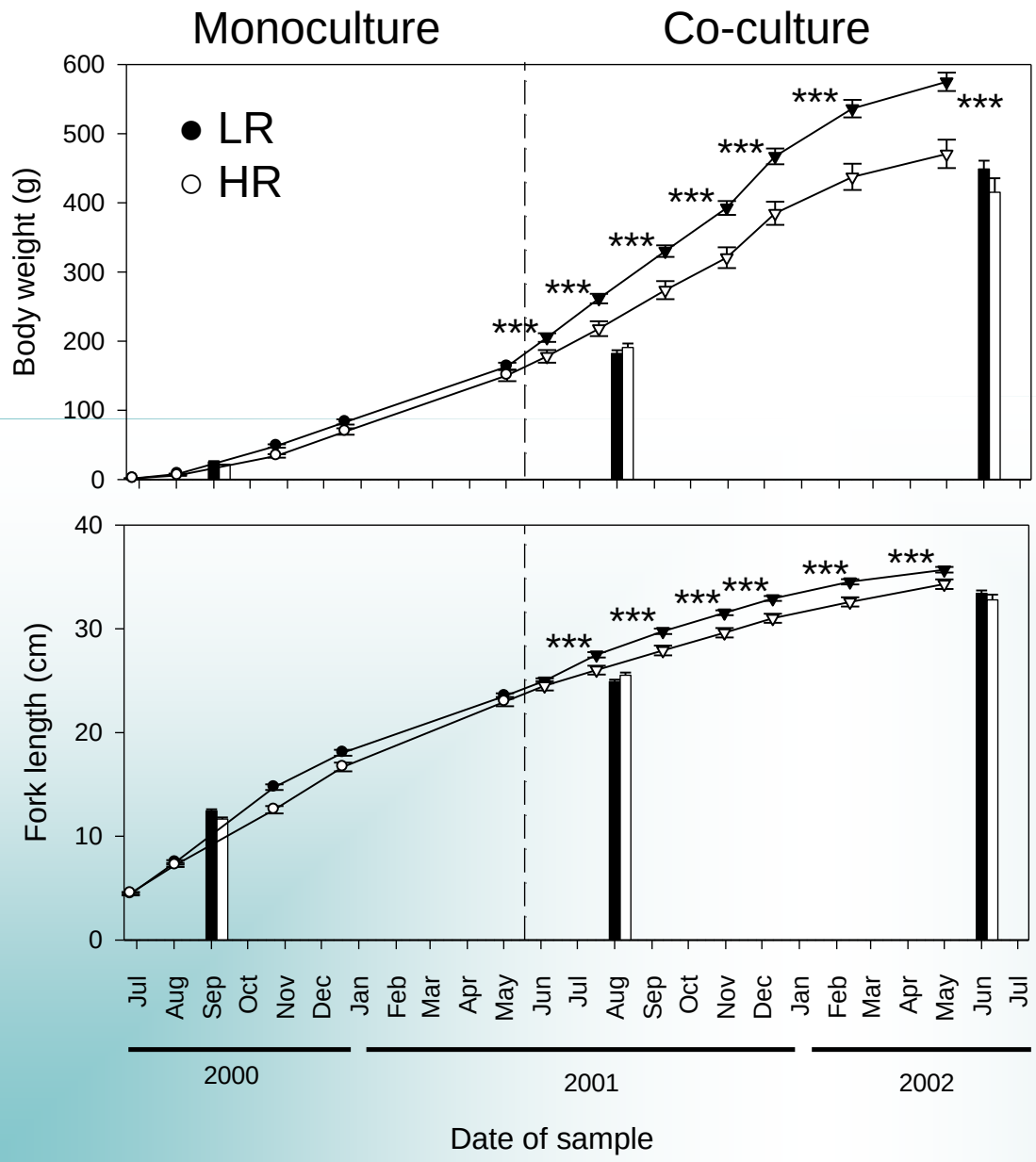
Better egg quality?

Higher survival of fry?

Flesh quality? – currently under re-investigation

Immunocompetence? – challenge results ambiguous

But - there is an additional complication.....



Performance of HR & LR lines :

Growth (F2)

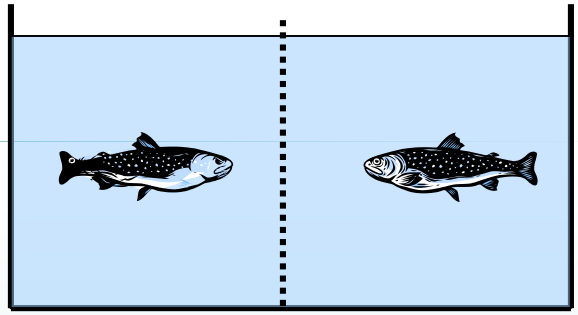
Monoculture:
HR = LR

Co-culture
HR < LR

Why?

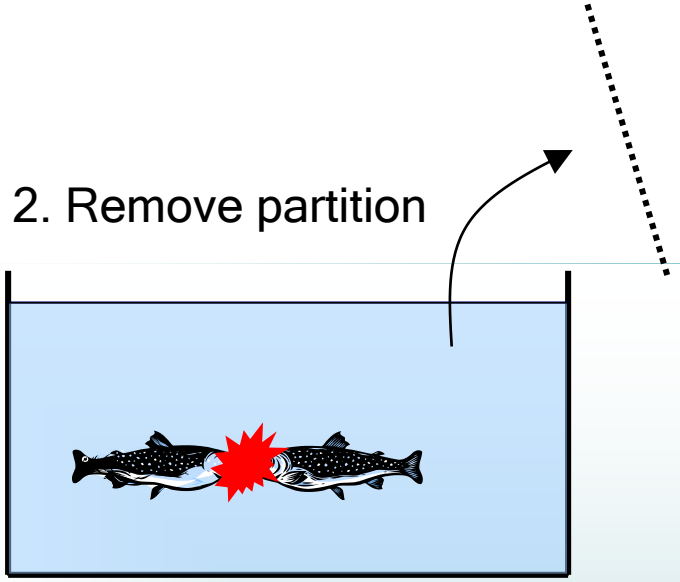
Behaviour of HR & LR lines (F2)**:**

Tendency for dominant/subordinate behaviour can be assessed in paired contests

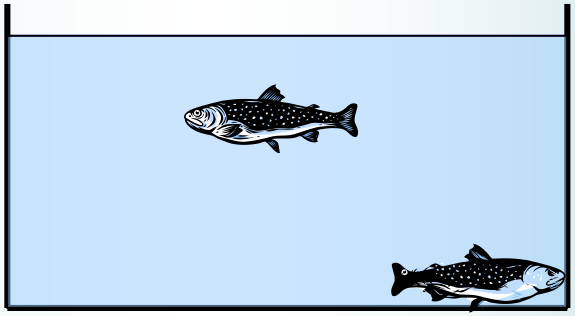


1. Isolate and acclimate

2. Remove partition



3. Fish assume dominant or subordinate status



Behaviour of HR & LR lines (F2):

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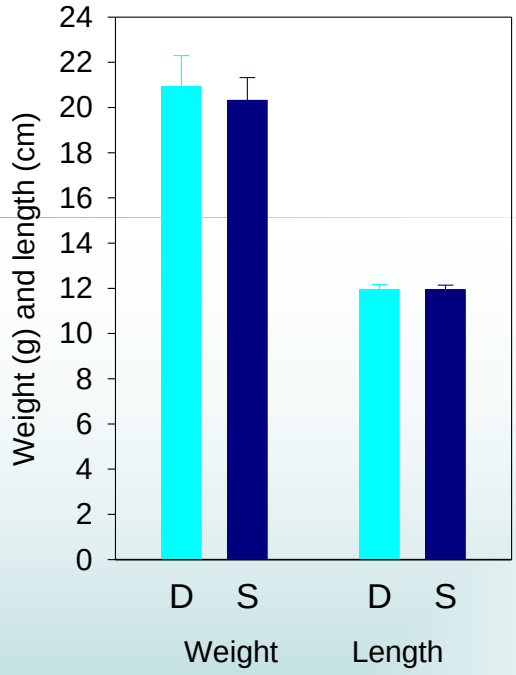
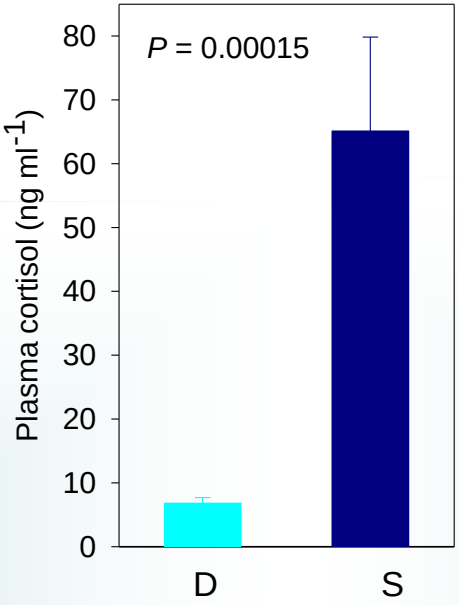
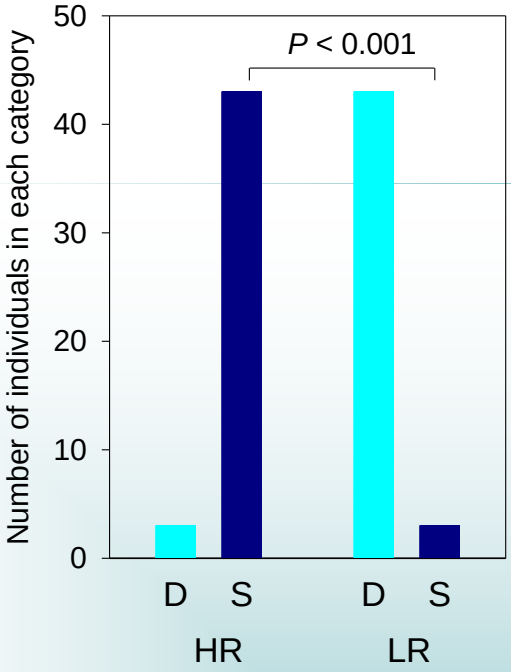
The outcome of paired contests between size-matched HR and LR fish







In 46 contests, LR was dominant in 43

There is an association between stress responsiveness and behaviour in the selected lines

- In co-culture LR trout grow > HR trout

  = food acquisition / aggression / competitive ability?

- In dyadic contests, LR fish are consistently dominant, HR are consistently subordinate

- Behavioural and physiological stress responses are controlled by common neuroendocrine signalling systems, e.g. monoamines, CRF

Two stress "coping styles" co-exist in animal populations (coping strategy, 'personality')

Coping styles:

‘***A coherent set of behavioural and physiological stress responses, which is consistent over time and which is characteristic to an individual, or a group***’

Koolhaas et al. (1999). Coping styles in animals: current status in behavior and stress-physiology. *Neurosci. Biobehav. Rev.* 23, 925-935.

Coping styles: pro-active & reactive (or passive)

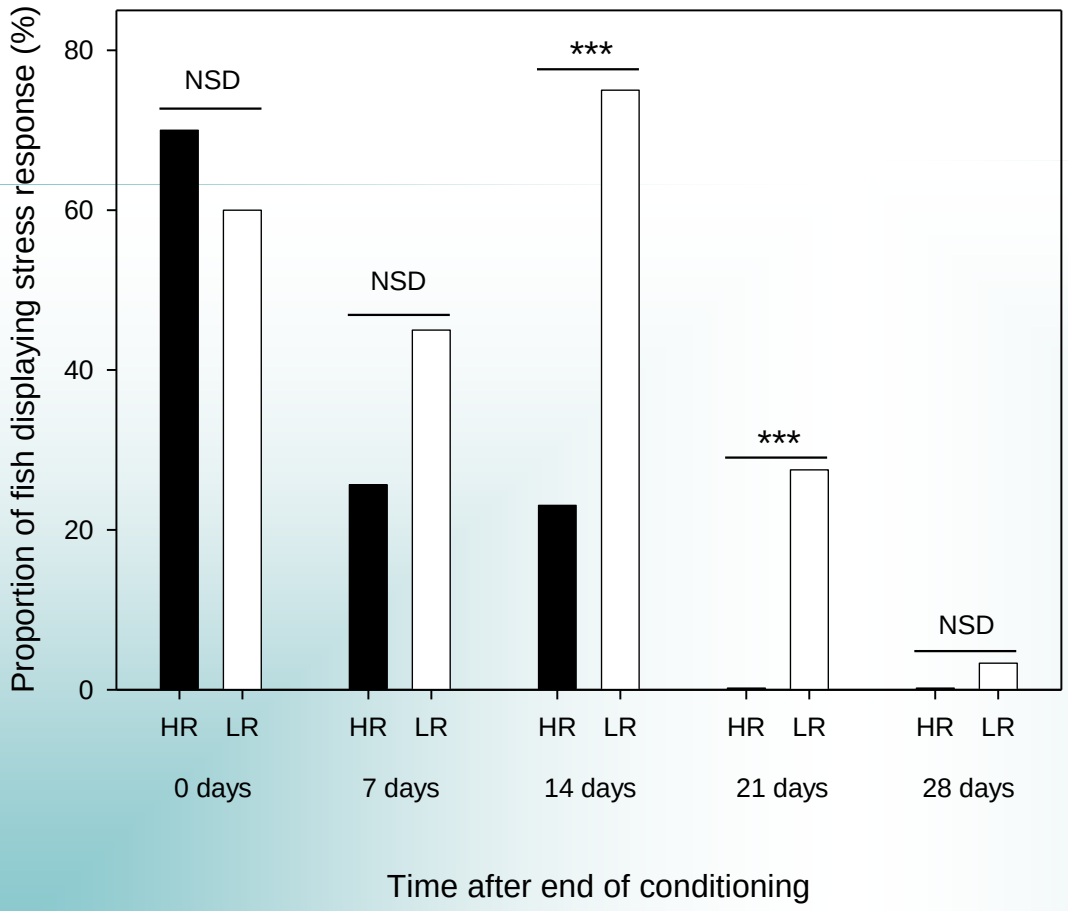
	Pro-active (=LR?)	Reactive (=HR?)
Corticosteroids	Low	High
Sympathetic activity	High	Low
Brain catecholamines	High	Low
Aggression	High	Low
Locomotor activity	High	Lower
Copes with novelty	Quickly	Slowly

Active (or pro-active) coping style: *'fight or flight' response*

Passive (or reactive) coping style: *conservation-withdrawal response*

Cognitive differences between the lines

Extinction of a conditioned response is delayed in LR fish



US – partial emersion
CS – water off

Conditioning = paired CS-US for 18 days

CR acquired in 12 days

Differences between HR & LR:

- *in learning/memory consolidation*
- *in consolidation/retrieval*
- *or at time of retrieval*

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CONCLUSION

Selection on a single endocrine trait results in phenotypes with distinct

physiological, behavioural and cognitive differences

These equate to “coping styles” and complicate the outcome of selection

Under aquaculture conditions LR is preferable to HR

But unclear (yet) whether LR is preferable to random-bred

FUTURE:

Outcomes of current QTL investigation (Aquafirst programme)

- *Marker assisted selection? Large scale trials?*

Continuation of lines and associated investigative work in Norway/Denmark

- Focus on aquaculture/behaviour interface e.g. reduced feed waste in LR lines following transfer

Final question – Should we ignore the magnitude of the response and focus instead on the trigger threshold?