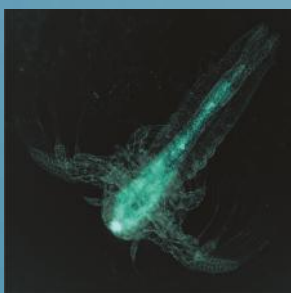
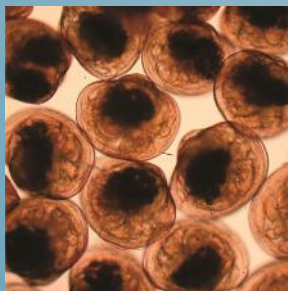
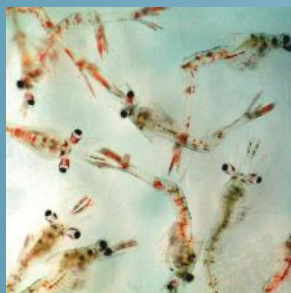
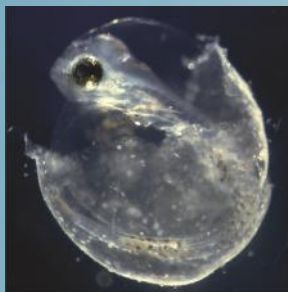


larvi 2013

6th fish & shellfish larviculture symposium

The long term effects of DHA supplementation
in live feed for first feeding pike perch larvae
on development of neural tissue
and behavioral responses

Ivar Lund



ghent university, belgium, 2-5 september 2013

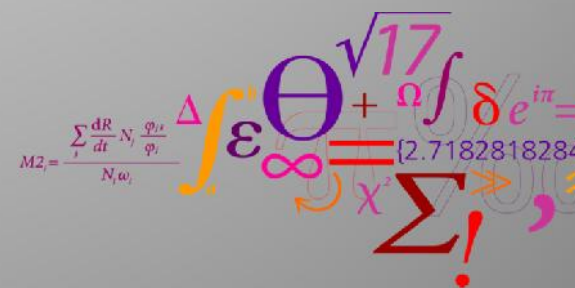


Importance of DHA for first feeding pike perch (Sander Lucioperca) larvae

- Influence on behavioural responses

Larvi – Ghent
2-6 Sept. 2013

Ivar Lund
Erik Høglund
Peter V. Skov





Challenges in production of pike perch fry

- Cannibalism
- Quality of fry, and survival rate
- High stress sensitivity



Causes

- Lack of breeding programme
- Environmental and nutritional issues
- **Optimal nutritional requirements not known**
(lipids, fatty acids, amino acids, vitamins etc.)

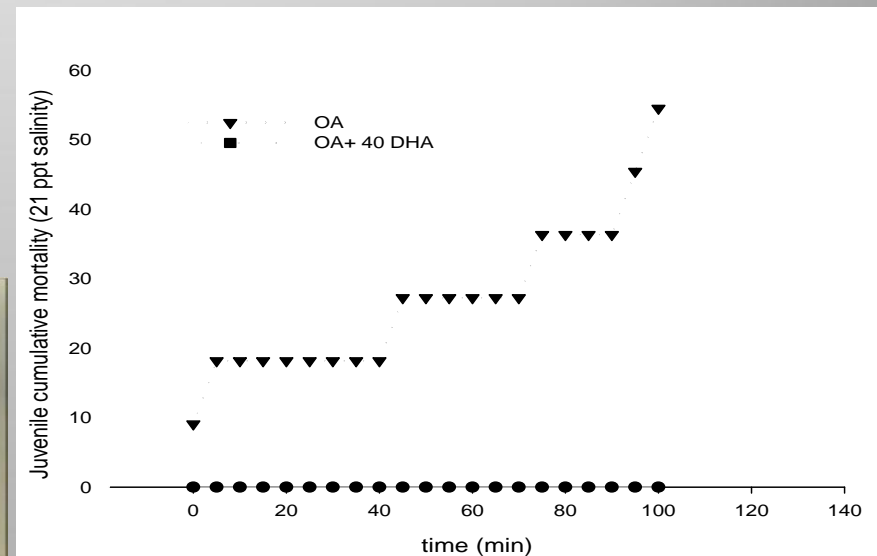
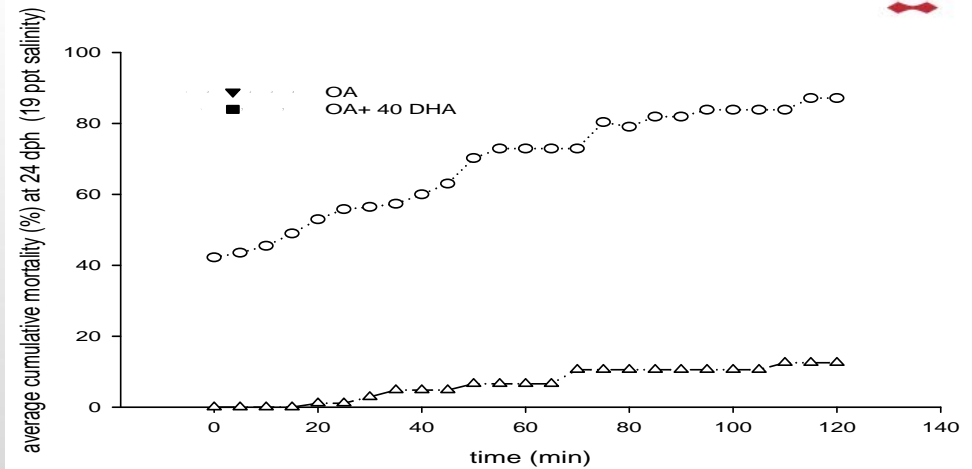


Influence of DHA on stress and neural development in pike perch larvae

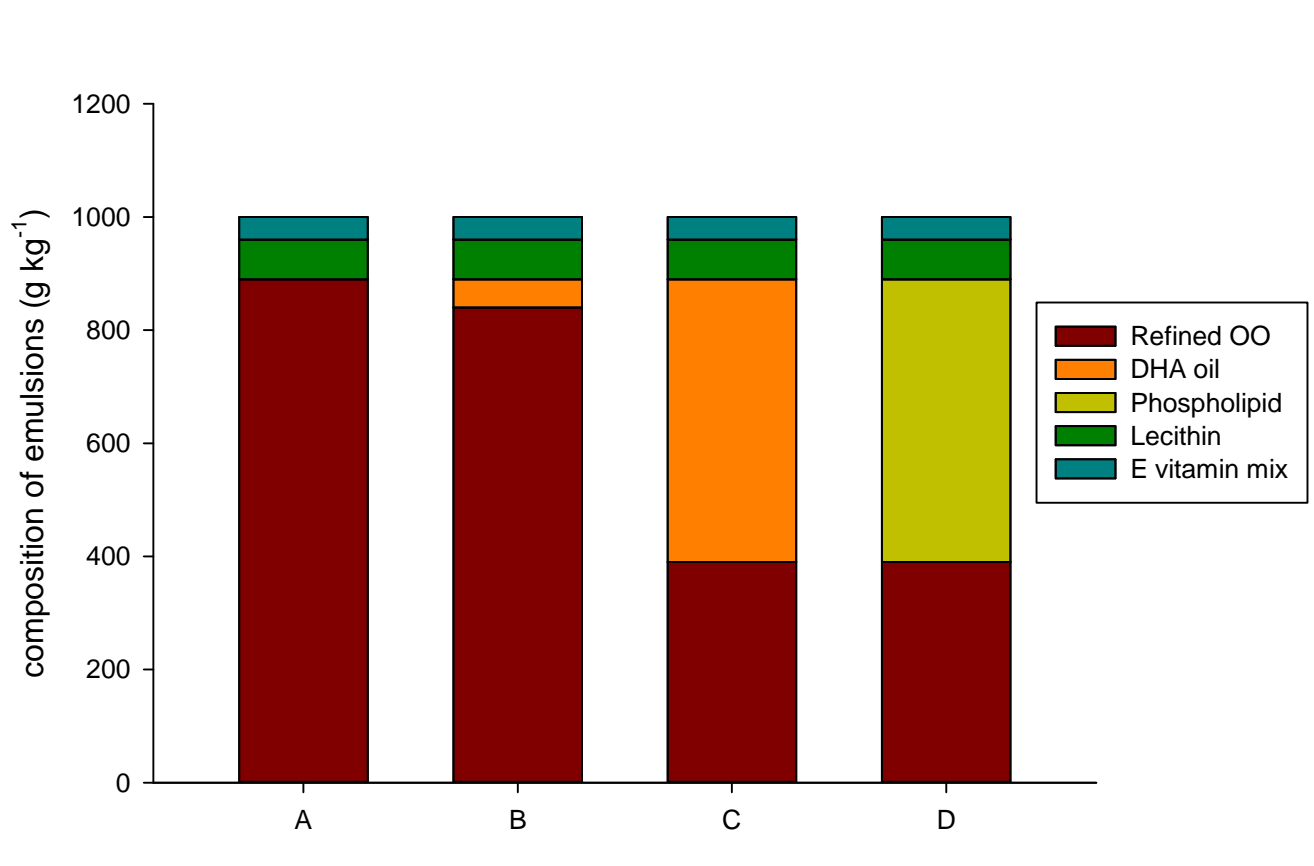
The developing brain may be vulnerable to DHA deficiency.

Neurologic processing,
Synapse formation, Neurite outgrowth,
Myelination, Neurotransmitter secretion
Neurological function.

Low brain DHA levels during development can be restored to normal by subsequent n-3 fatty acid supplementation, but will physiological functions be affected ?



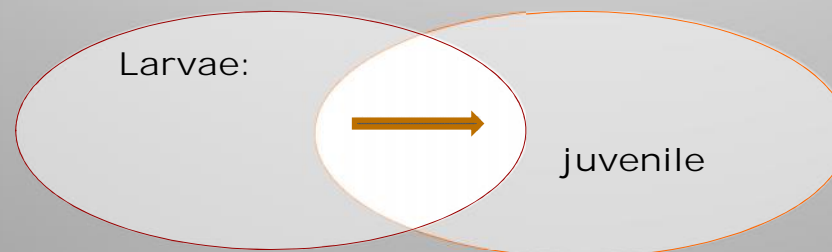
Experimental set up:



OO: refined olive oil (0.79 kg 18:1 kg oil⁻¹)
 DHA oil: INCROMEGA, CRODA, (0.51 kg DHA Kg oil⁻¹)
 Phospholipid oil (PL): fish oil rich in phospholipids (0.44 kg phospholipid kg oil⁻¹)

Experimental set up:

- Behaviour of larvae (28-30 dph) in presence and absence of a simulated predator
- Fast-escape response (avoidance behaviour) of larvae (28 dph) and juveniles (+120 dph) to a mechanosensory stimulus
- Long terms effects on learning ability and stress responsiveness by maze tests and cortisol analyses

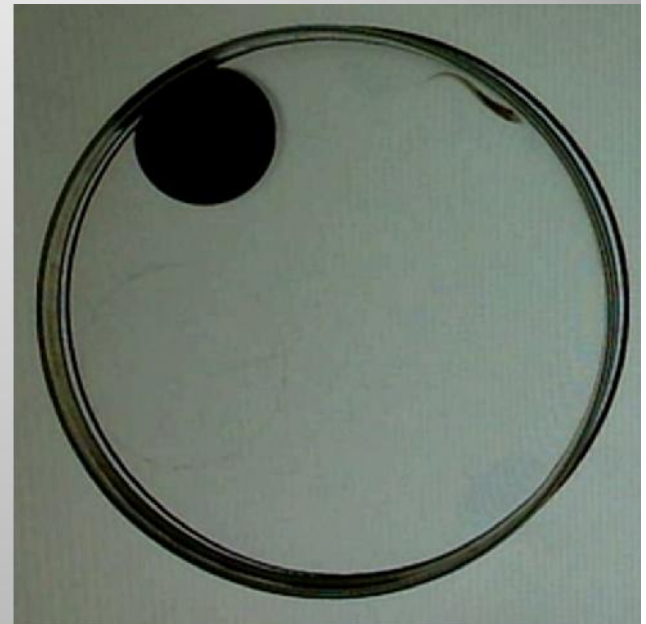
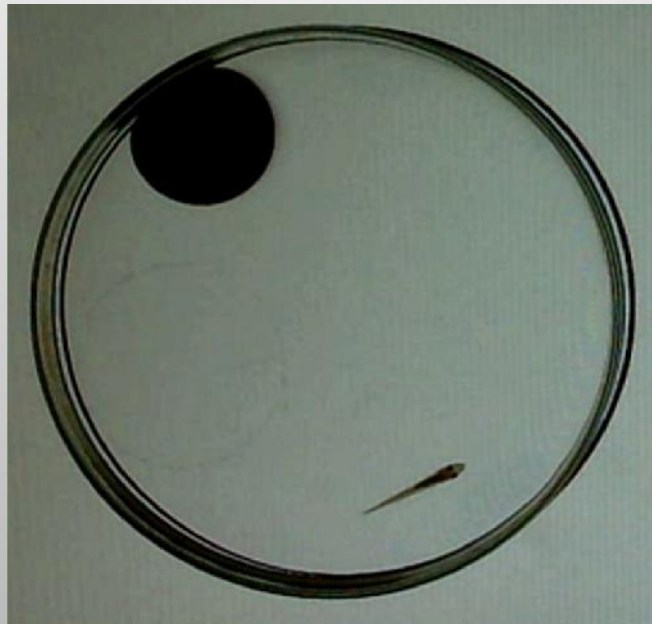
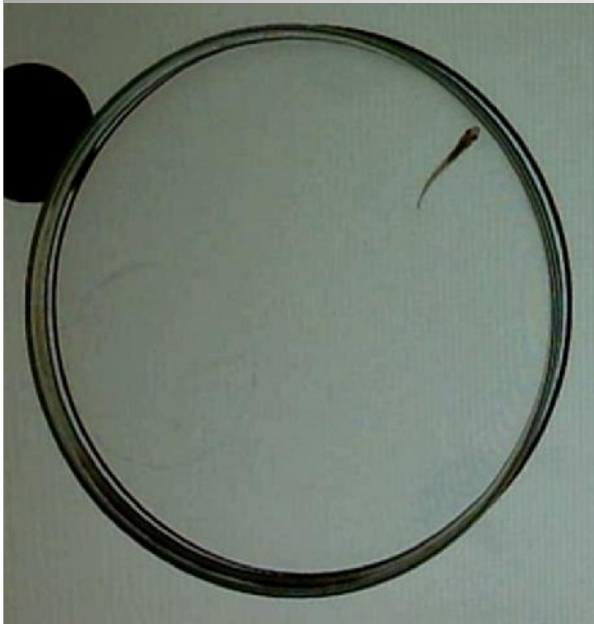


Physiological implications of early larvae FA nutrition:
Short & long term consequences on welfare, cognitive behaviour;
stress

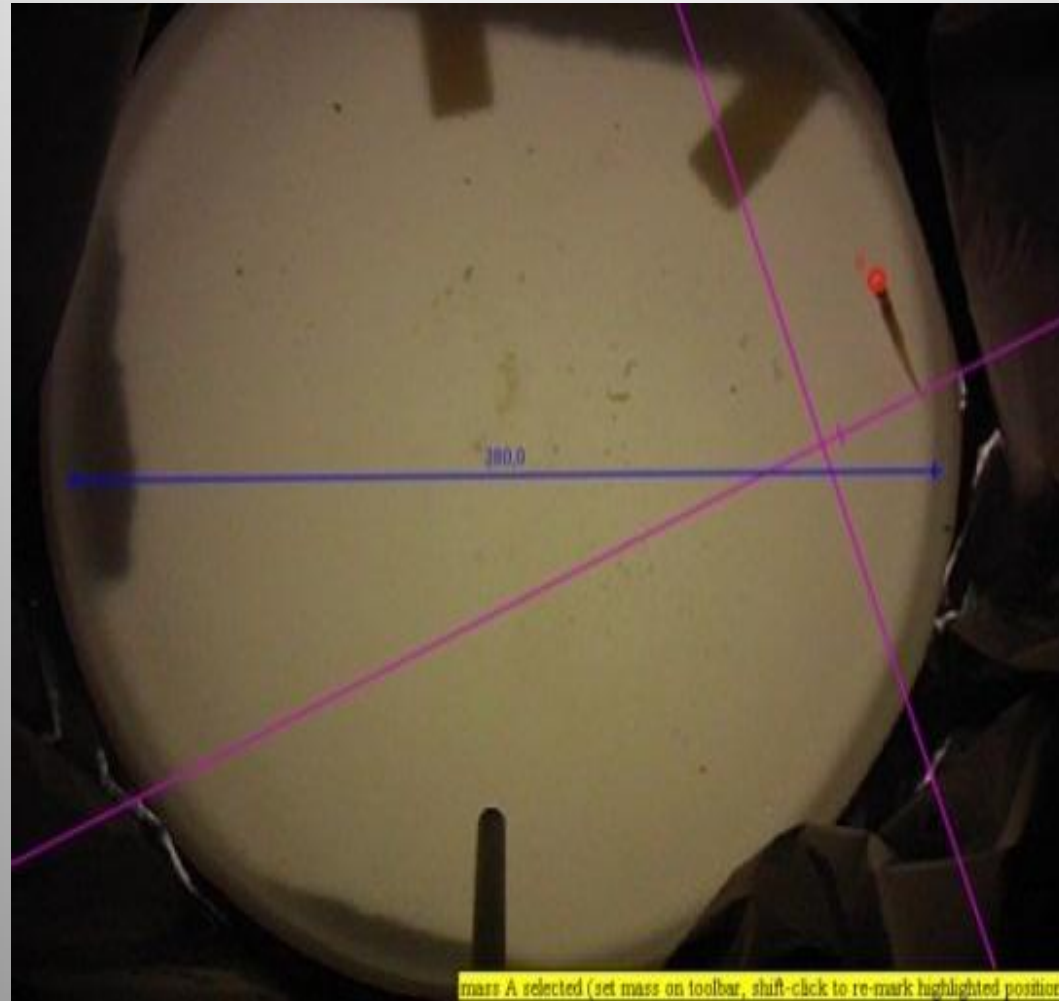
Experimental set up:



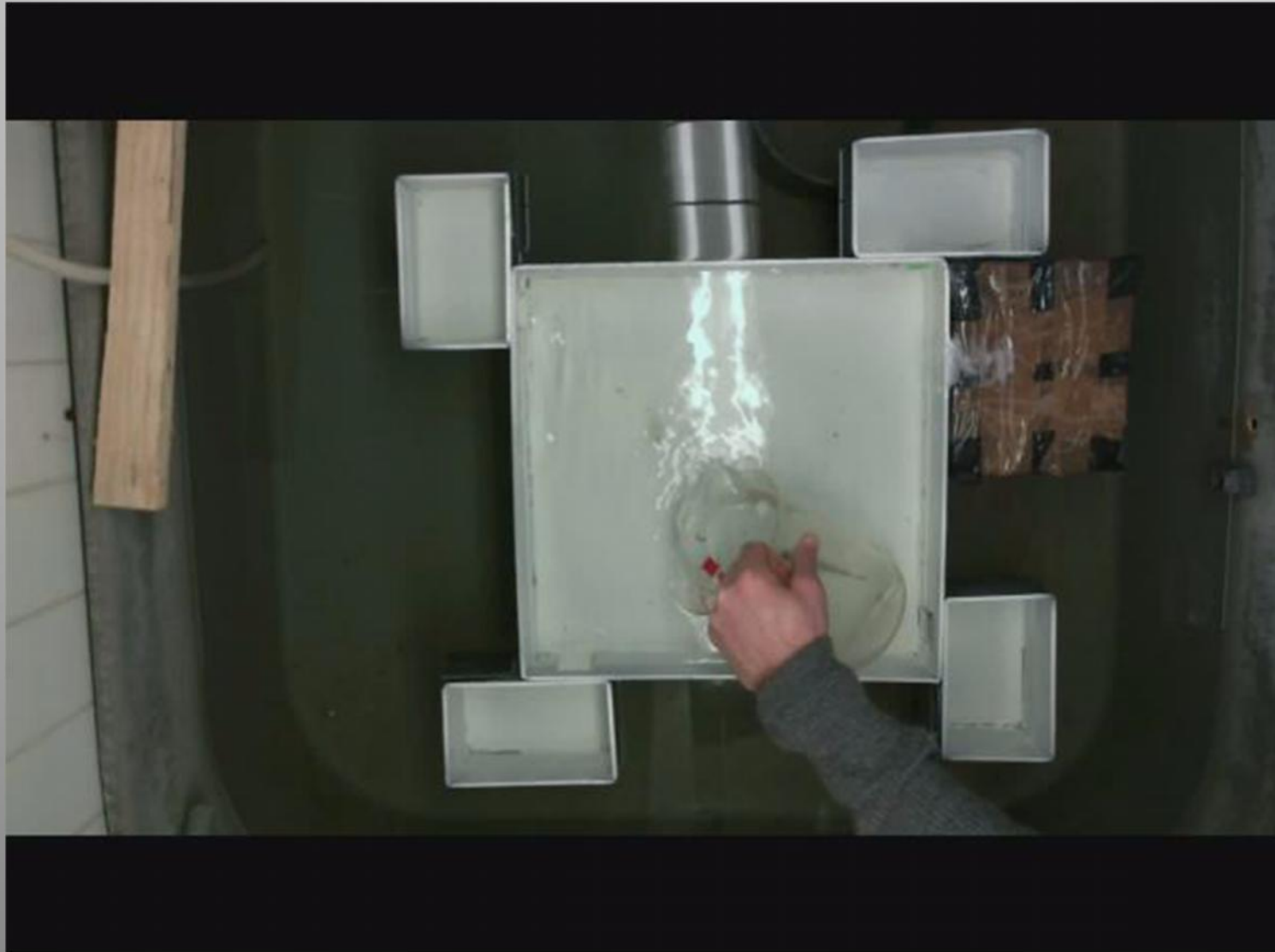
Behaviour of larvae in absence and presence of a simulated predator



Fast/ escape (avoidance) test for larvae & juveniles



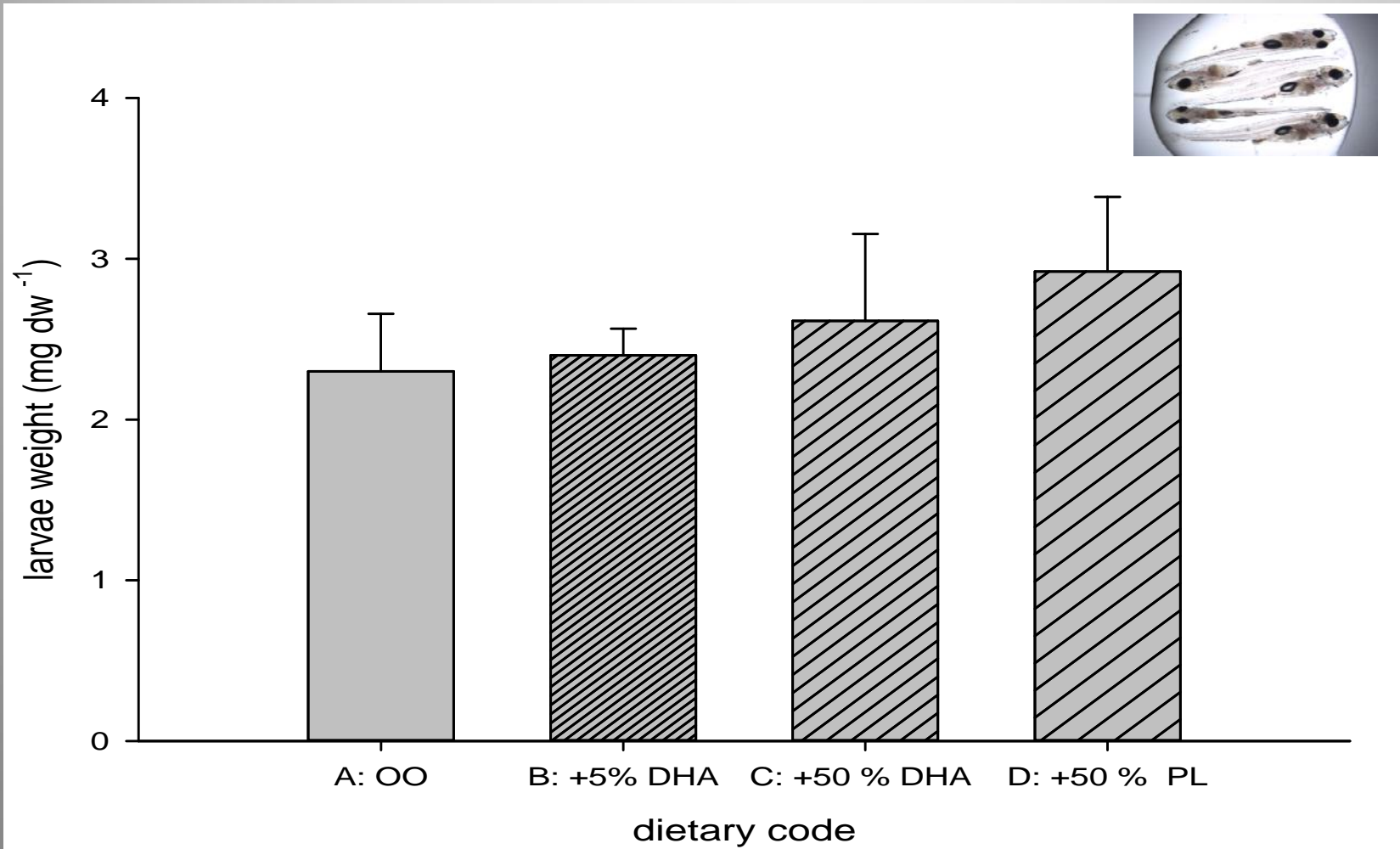
Learning ability – maze test



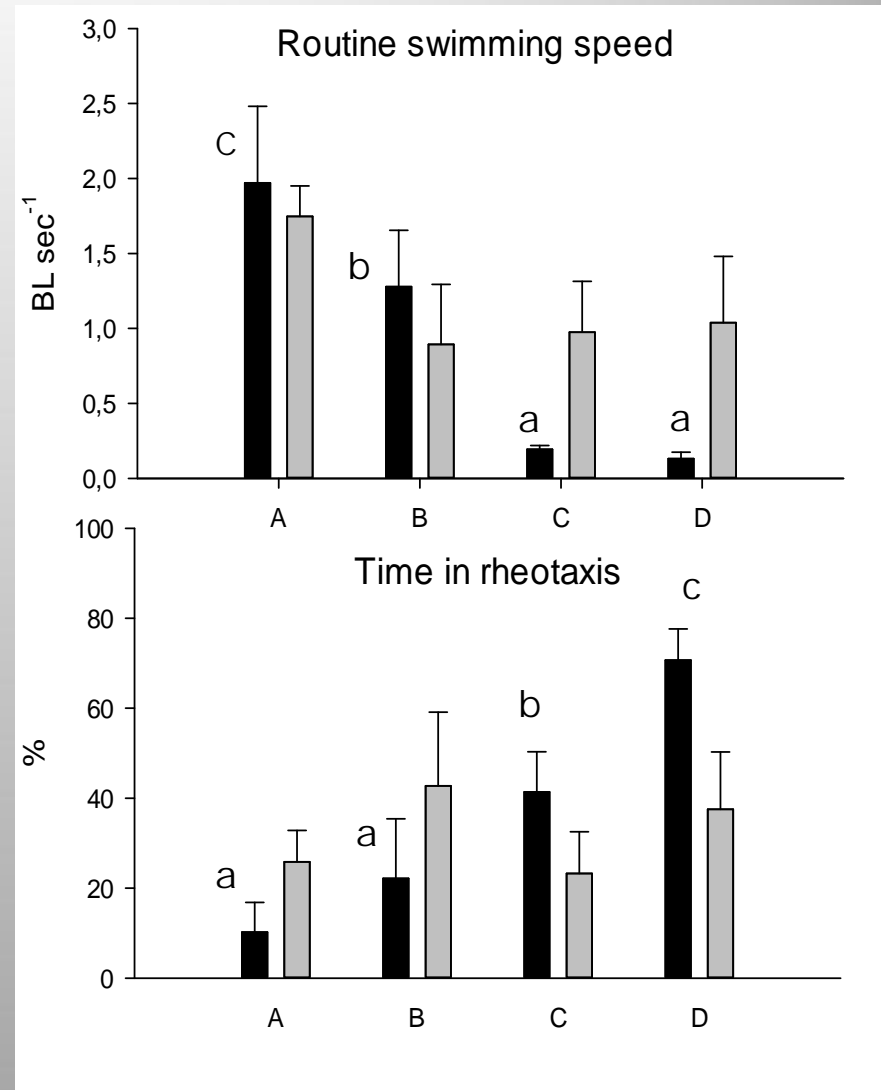
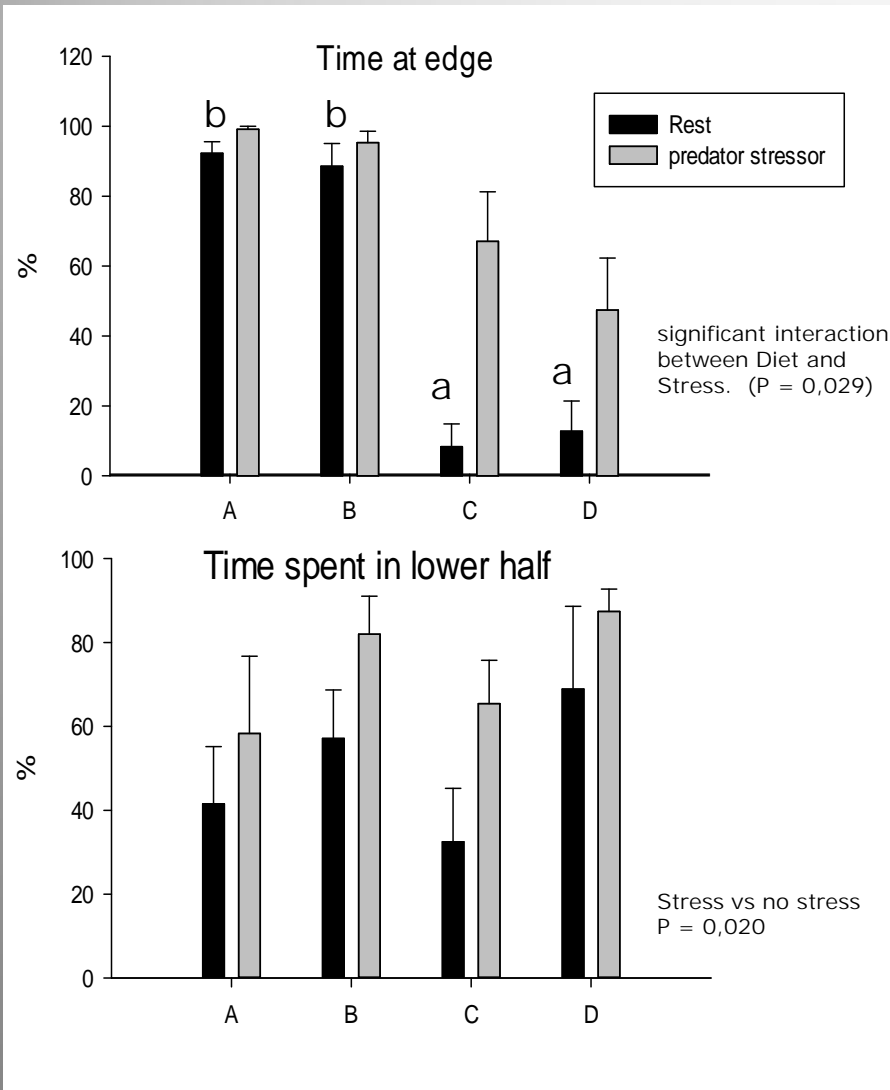
Larvae FA composition 27 dph:

Dietary inclusion:	A: 890 g OO	B: 840 g OO 50 g DHA oil	C: 390 g OO 500 g DHA oil	D: 390 g OO 500 g PL.
Larvae FA comp (% total) (27 dph):				
16:0	16.9±0.4	17.0±0.3	15.9±1.0	15.9±0.4
18:0	8.3±0.3	8.6±0.3	8.9±0.3	7.8±0.3
SFA	26.6	27.2	26.6	25.5
16:1 n-7	1.6±0.1	1.5±0.1	1.3±0.1	2.1±0.4
18:1 n-9	34.1±1.4^d	31.9±1.5^c	24.0±1.1^a	28.1±1.0^b
MUFA	38.6 ^d	36.4 ^c	29.6 ^a	34.1 ^b
18:3 n-6	0.4±0.0	0.4±0.0	0.3±0.0	0.3±0.0
20:4 n-6	0.9±0.1 ^a	1.0±0.1 ^a	1.5±0.1 ^b	1.0±0.1 ^a
n-6 PUFA	7.9	7.5	6.7	6.4
18:3 n-3	18.6±0.3	18.9±0.5	19.2±0.7	17.2±0.3
20:5 n-3	2.6±0.2 ^a	3.3±0.3 ^a	5.4±0.3 ^b	6.4±0.4 ^c
22:6 n-3	3.5±0.6^a	4.6±0.1^b	10.3±1.3^c	8.5±1.4^c
n-3 PUFA	26.1 ^a	28.1 ^b	36.2 ^d	33.3 ^c
DHA/EPA	1.4	1.4	1.9 ^b	1.3
n-3 /n-6	3.3	3.7	5.4	5.2

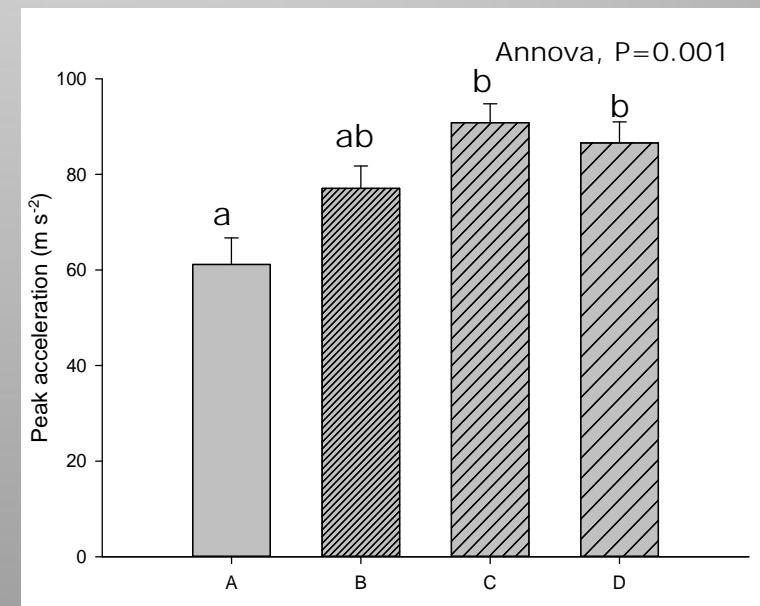
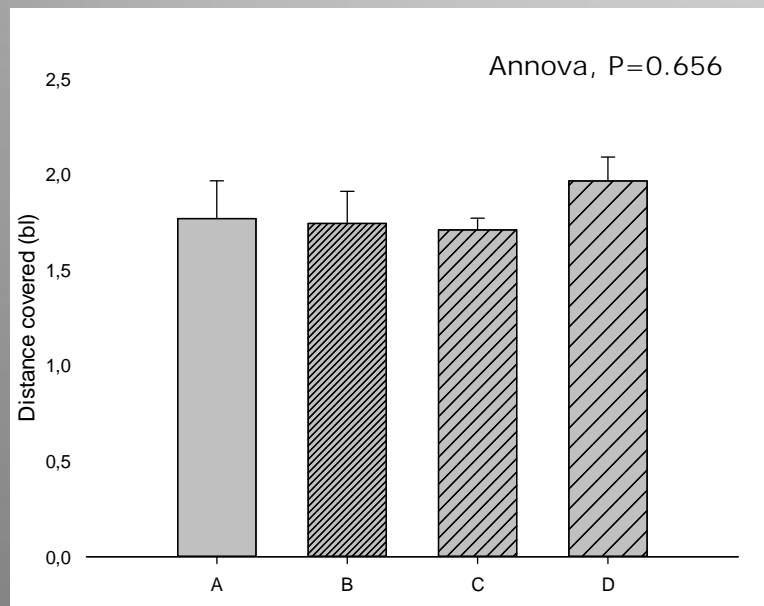
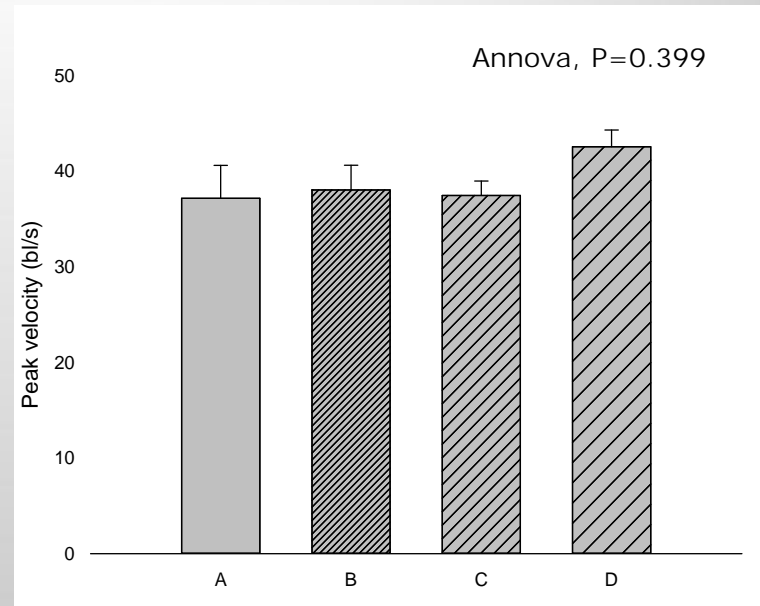
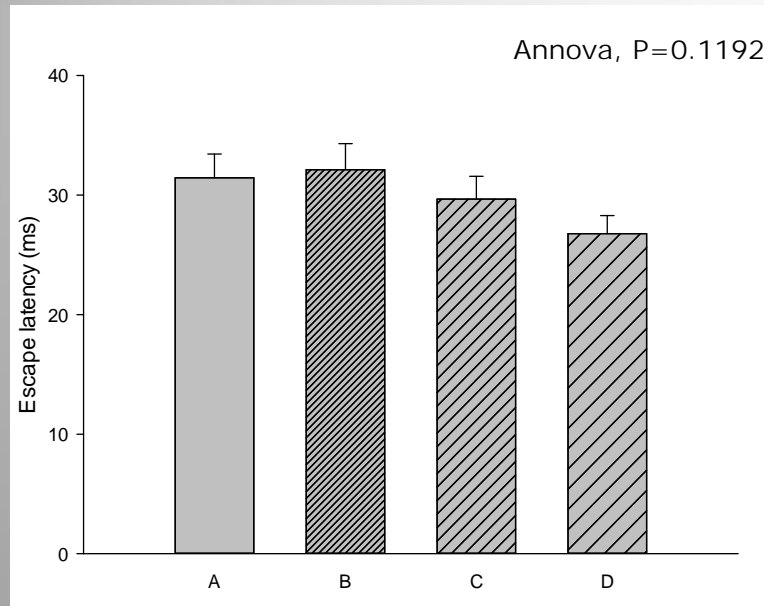
Larvae growth until 27 Dph



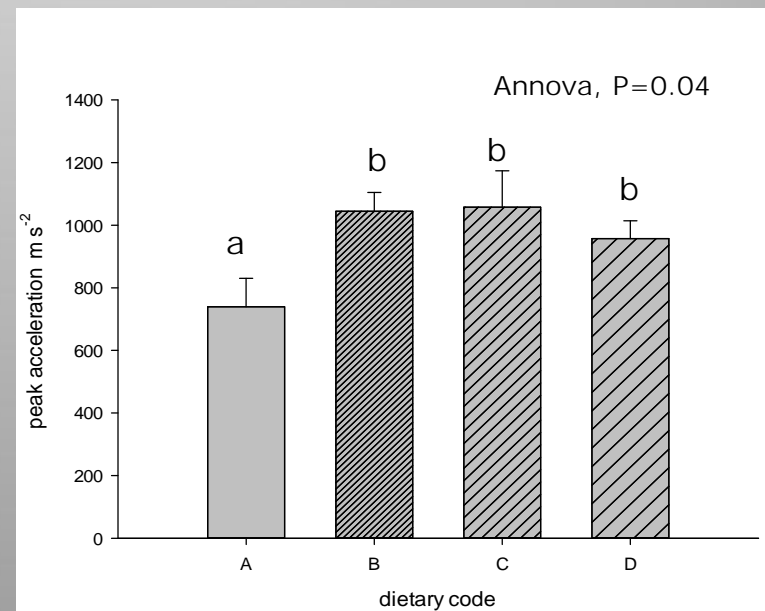
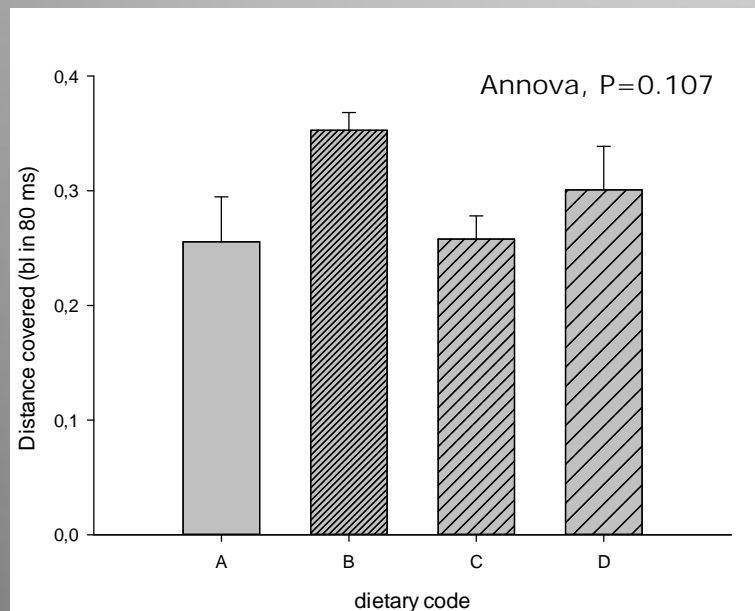
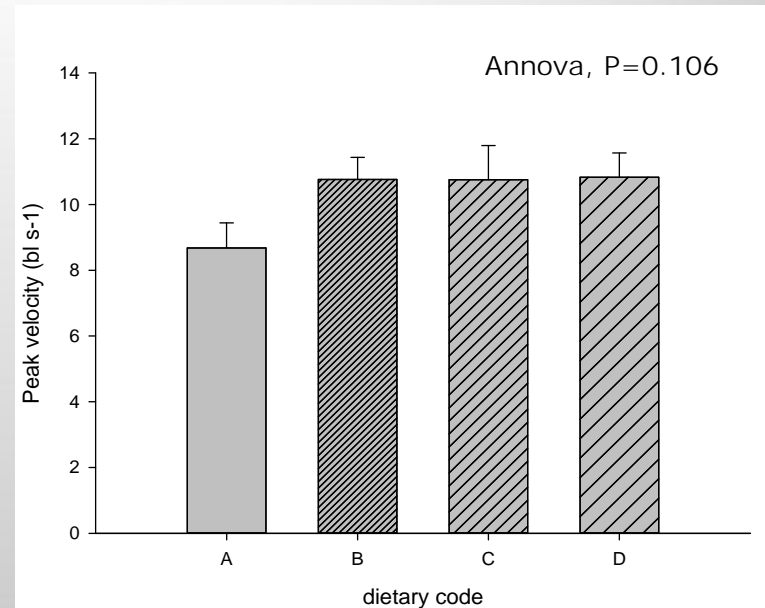
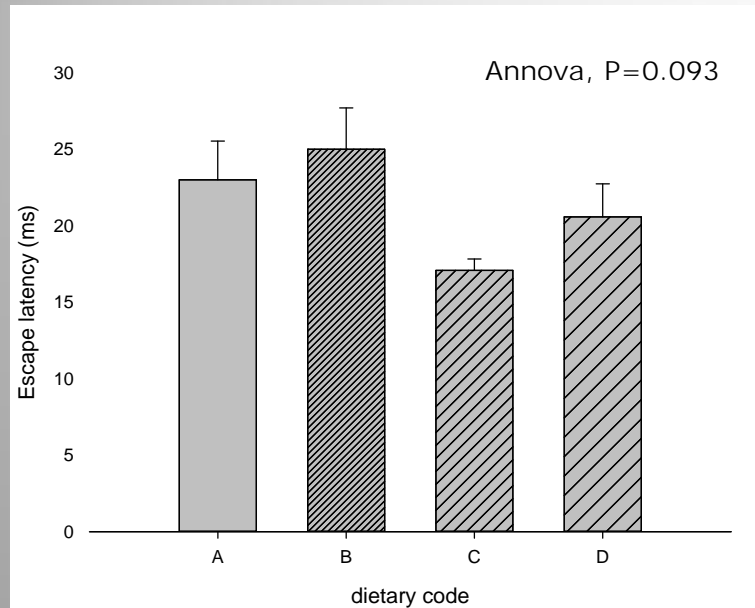
Larvae behaviour in absence and presence of a simulated predator



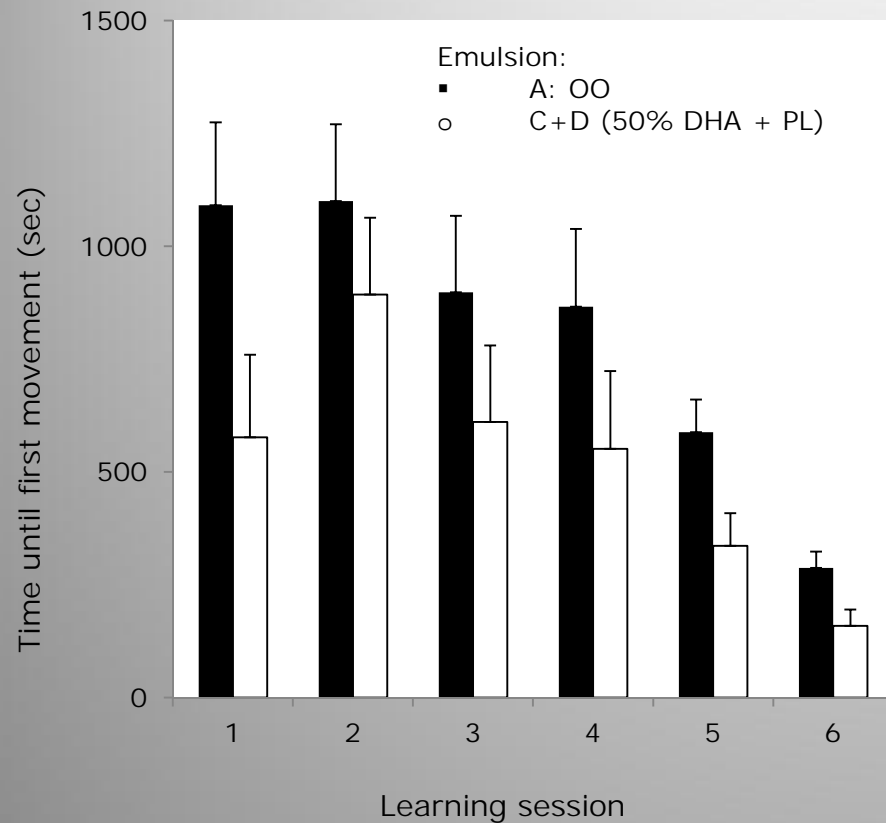
Larvae (avoidance) escape response



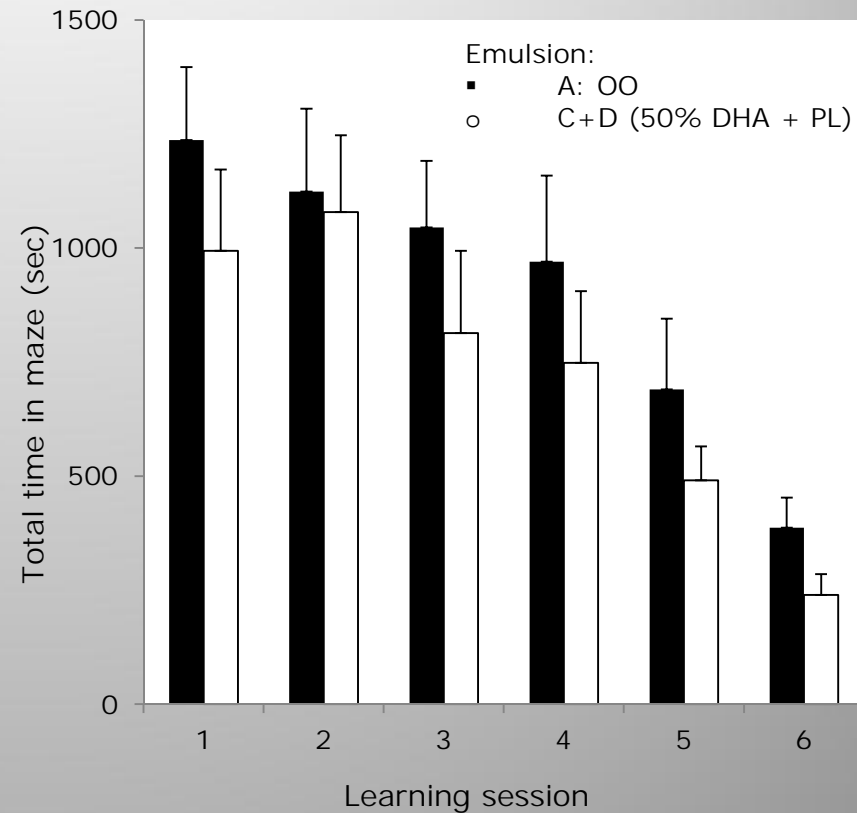
Juvenile (avoidance) escape response



Learning ability and stress responsiveness in maze tests

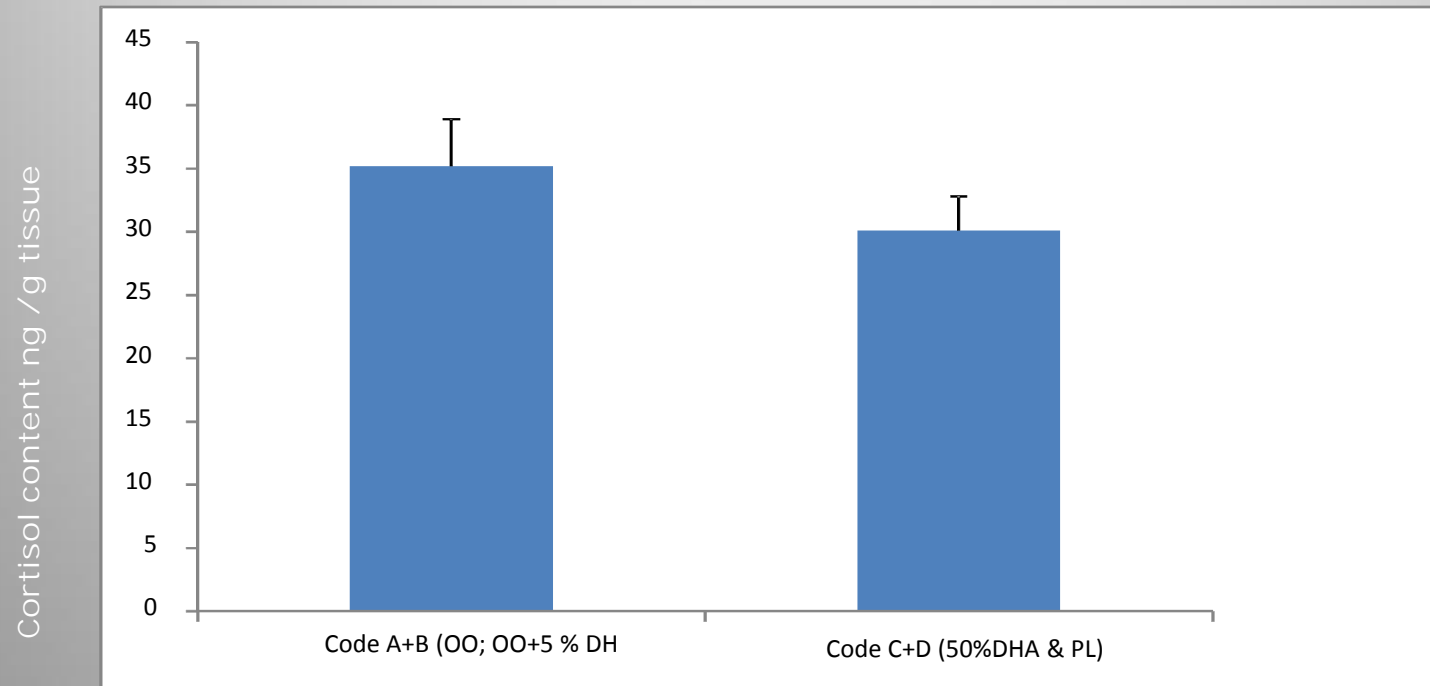


Two way repeated ANOVA
Learning session: $P < 0.001$
Fatty acid: $P < 0.05$
Fatty acid x Trail: 0.56



Two way repeated ANOVA
Learning session: $P < 0.001$
Fatty acid: $P < 0.20$
Fatty acid x Trail: 0.55

Cortisol levels in juveniles after test in maze



Significance: (P=0.25).

Conclusions

- Dietary DHA influences on larvae tissue composition and affects larvae stress responsiveness and behavior during rest or to an acute stressor.
- No apparent influence on growth or visual acuity.
- Peak acceleration for larvae and juveniles were significantly decreased by low dietary DHA concentrations.
- - n should be increased in such studies as individual variation is high.
- Juvenile learning ability in a maze was similar between groups, but "initial freezing time" higher in groups fed low levels of DHA as larvae.

- future studies

- How is stress and behavior linked to neurophysiology ?
 - a cross disciplinary approach including brain signaling systems as well as behavior and metabolic physiology.

- From an applied point of view, gene expression or protein levels of BDNF (i.e neurotrophic factor associated with learning) may be utilized as biological markers for stress coping ability and mental robustness in fish.



Aknowledgements

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Thanks to:

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University of Roskilde, Enspac for GC/MS analyses