OSMO_LUS
Osmoregulation in Salmon Lice

Anna Z Komisarczuk

23.01.2018

Aims

Fresh water treatment is one of the methods currently used to fight salmon lice infections

OSMO_LUS project:

- ➔ gain more knowledge about fresh water tolerance in salmon louse;
- ➔ investigate physiological changes in salmon louse triggered by fresh water exposure;
- ➔ identify the main players (genes/proteins) involved in osmoregulation in salmon louse;
- ➔ identify anatomical structures associated with osmoregulation;

Osmoregulation

OSMOREGULATION:

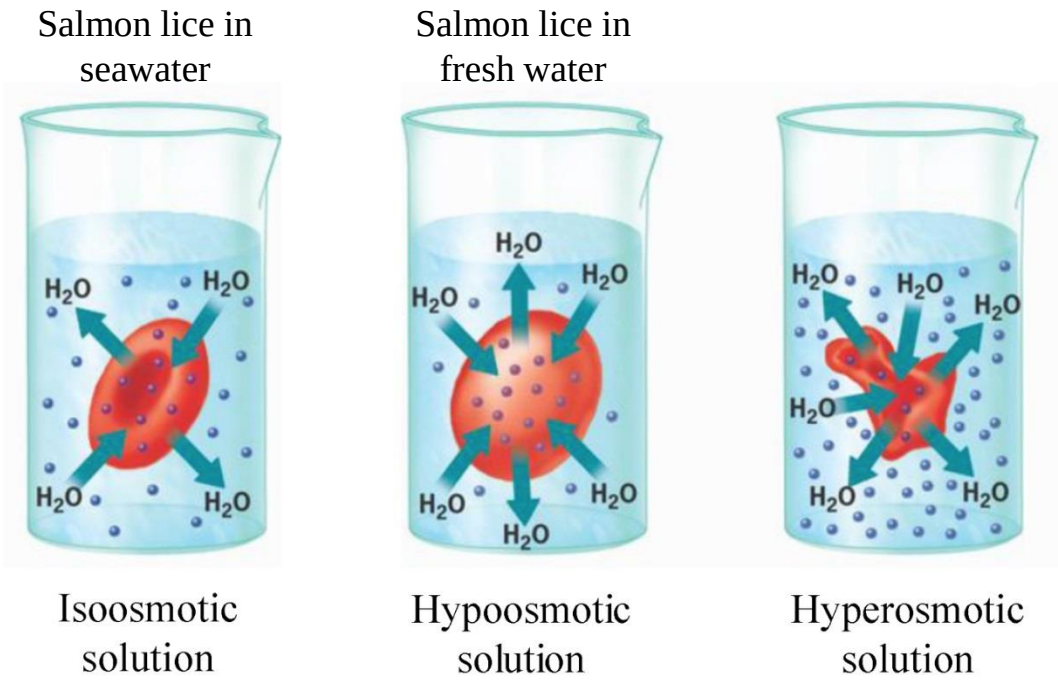
The ability to maintaining homeostasis with respect to solute (mainly salts) concentration and balance of water content.

- It is achieved by controlled movement of salts and water between internal fluids and the external environment through selectively permeable membrane.
- Homeostasis of cell/organism has often narrow limits;

Why?

Salts concentration in a external solution, determine movement of water across a selectively permeable membrane:

- **Isosmotic solution**: equal concentrations of water and salts – the movement of water is equal in both directions
- **Hypoosmotic solution**: lower salts and higher free water concentration – water movement to cell;
- **Hyperosmotic solution**: higher salts and lower free water concentration – water movement from cell;



Osmoregulation in Salmon Louse

Regulation of water transport through body/cell surface is **crucial for organisms living in water**.

Environmental solution has very high or very low salt content, often different than concentration of body and tissue fluids:

- Sea water: ≈ 34 ppt salinity = 1000 mOsm/l
- Fresh water: < 3 ppt salinity = 1 – 10 mOsm/l

Salmon louse (*Lepeophtheirus salmonis*) is **osmoconformer** – it is isoosmotic with the marine environment and in normal conditions does not regulate its osmolarity (the internal osmolarity of the salmon louse – 1000mOsm/l).

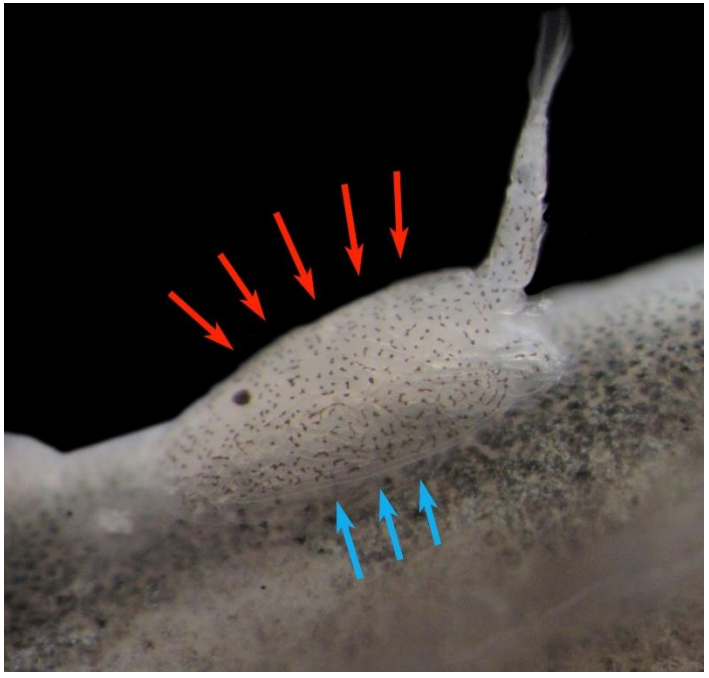
Osmoconformers have low tolerance of osmolarity change of their environment

Without the host:

➔salmon louse cannot tolerate substantial changes in external osmolarity and die within hours

When attached to its host:

➔ salmon louse is able to survive large fluctuations in external osmolarity, and control water influx in hypoosmotic environment by uptake of osmolytes from its host.

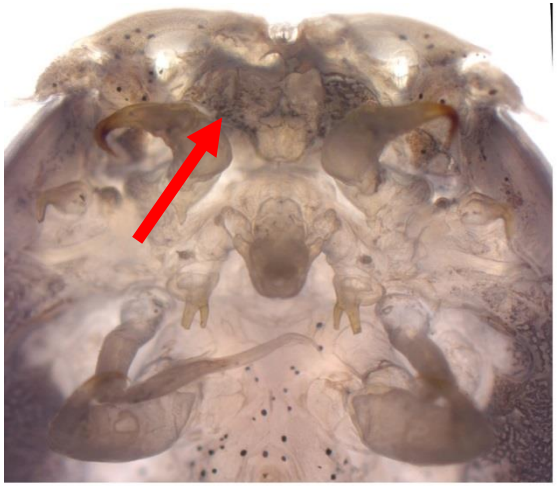


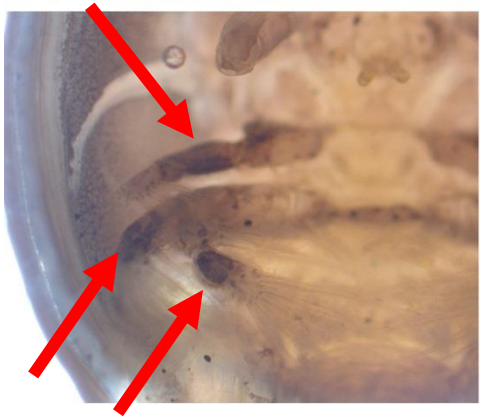
Ion Exchange/Transport Structures

Salmon louse lacks obvious osmoregulatory structures (usually gills).

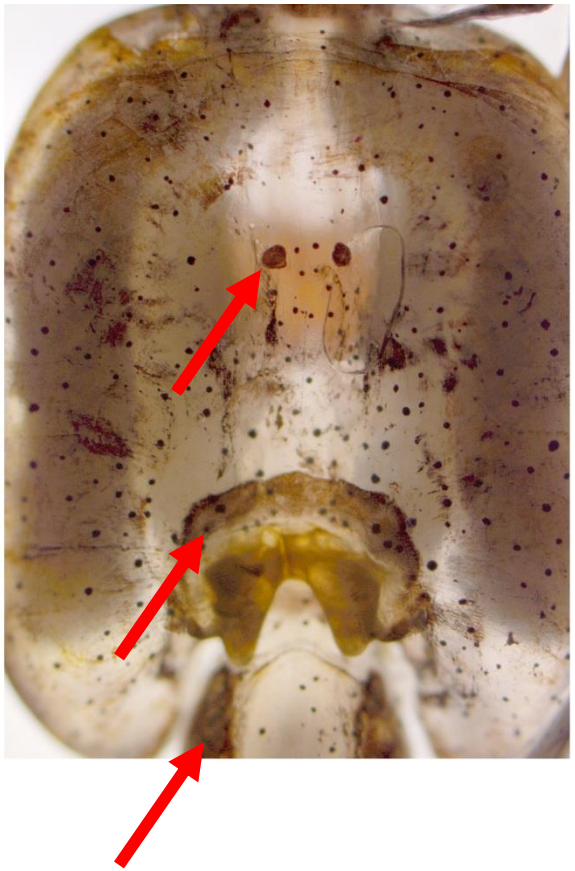
➔ Ion exchange/transport structures were identified in many locations on the animal body

Adult female





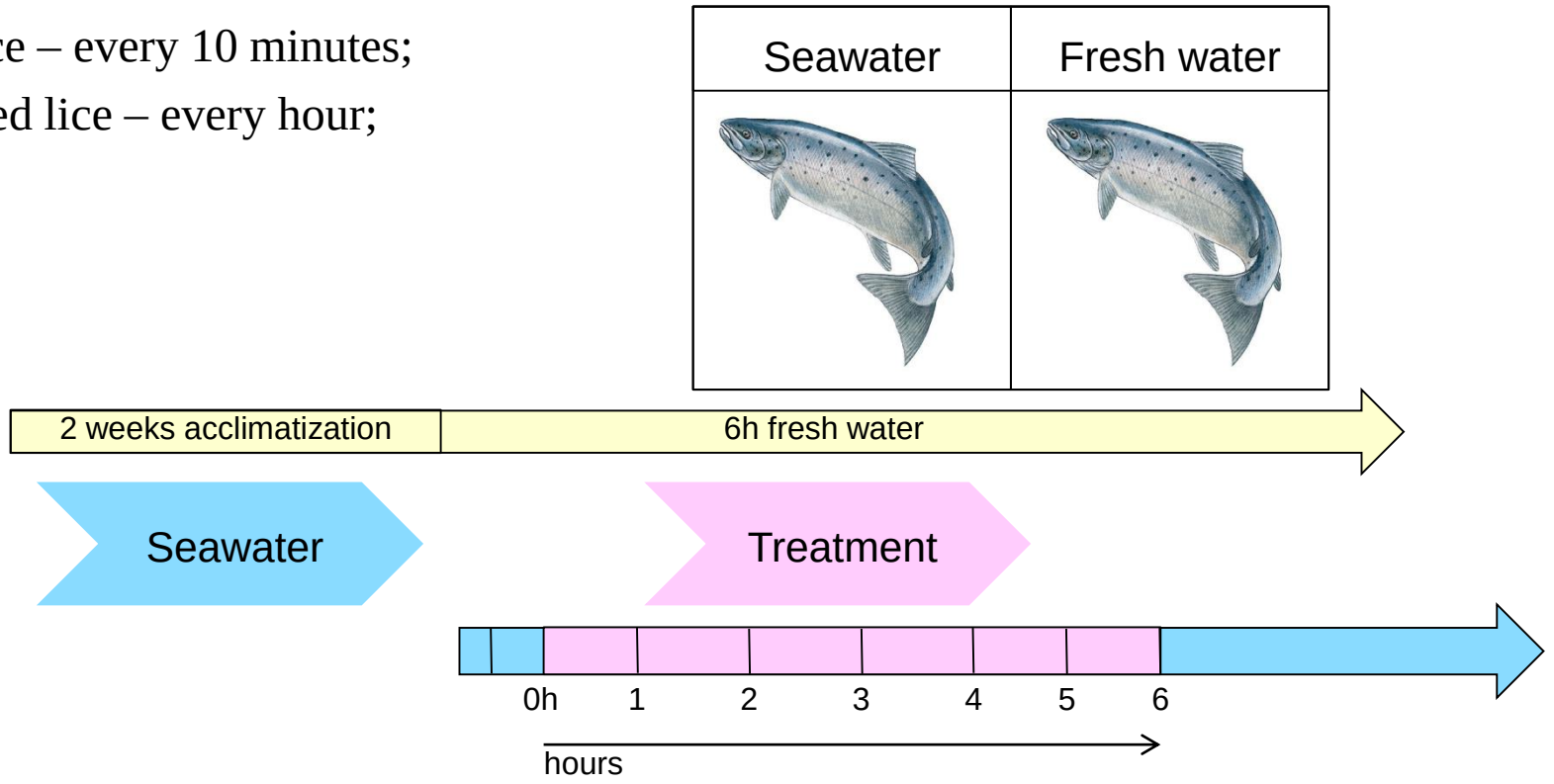
Crusalis organs



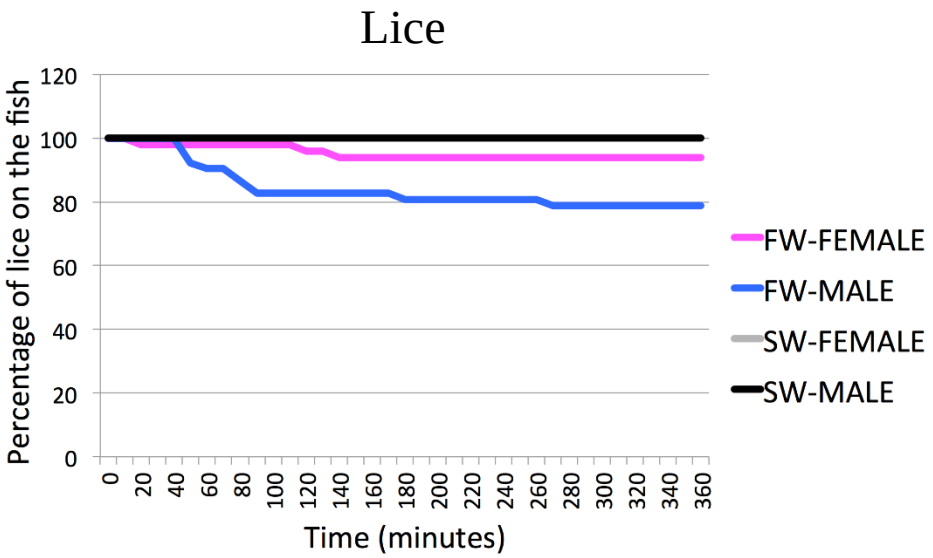
Copepodid

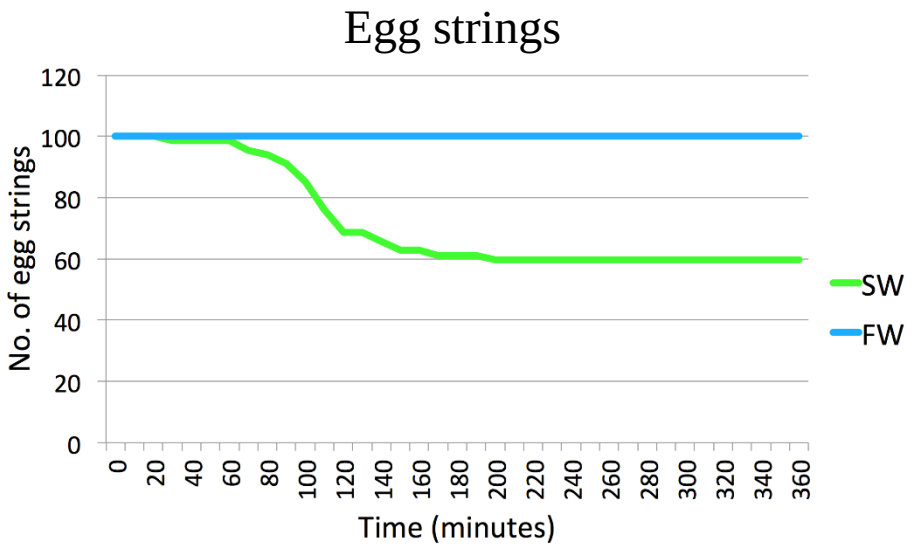
Fresh Water Effects on Adult Lice

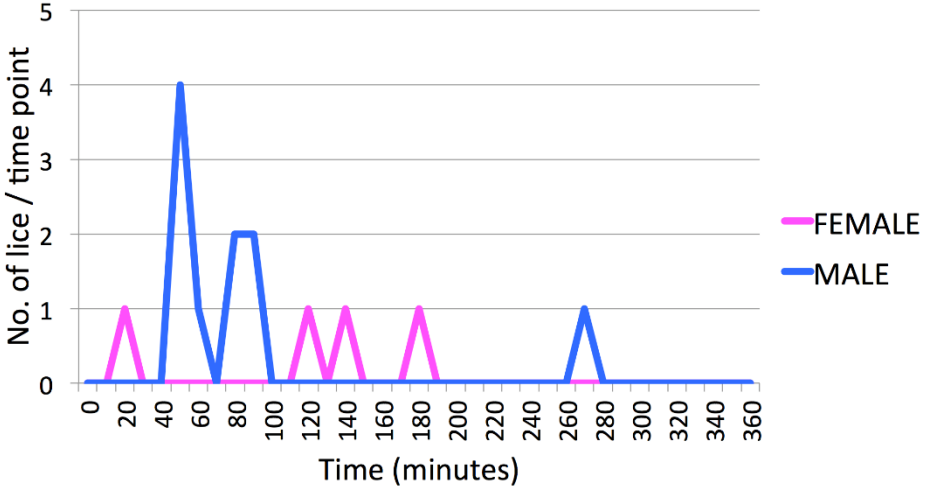
- Treatment time – 6h
- 2 groups: Fresh water and Seawater
- Sampling:
  - Lost lice – every 10 minutes;
  - Attached lice – every hour;

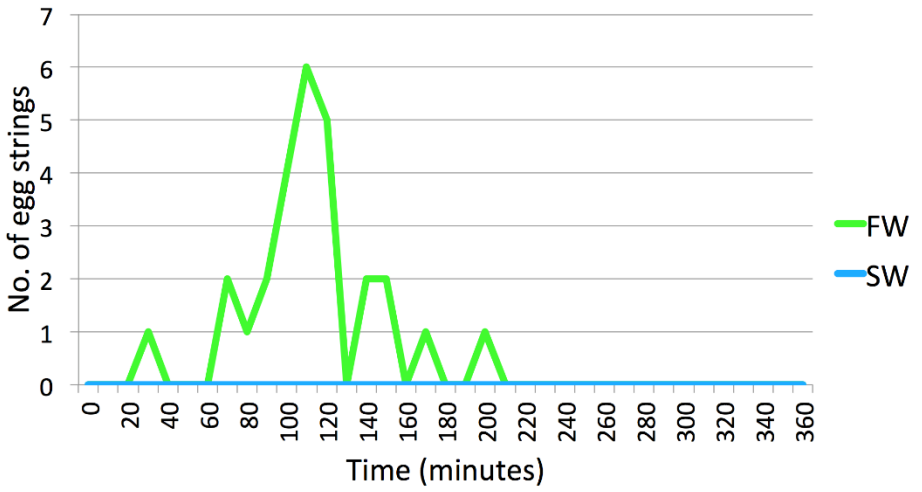


Fresh Water Effects on Adult Lice – Lice Loss





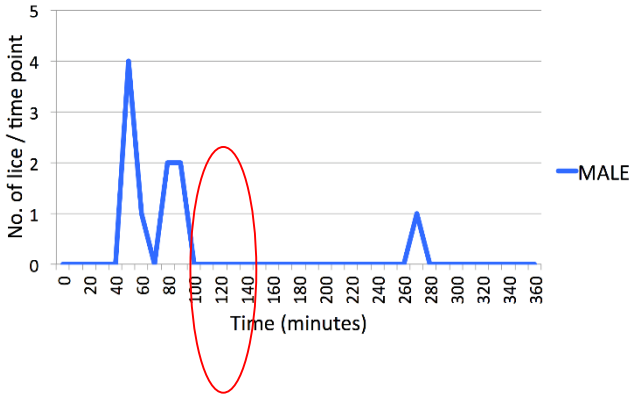




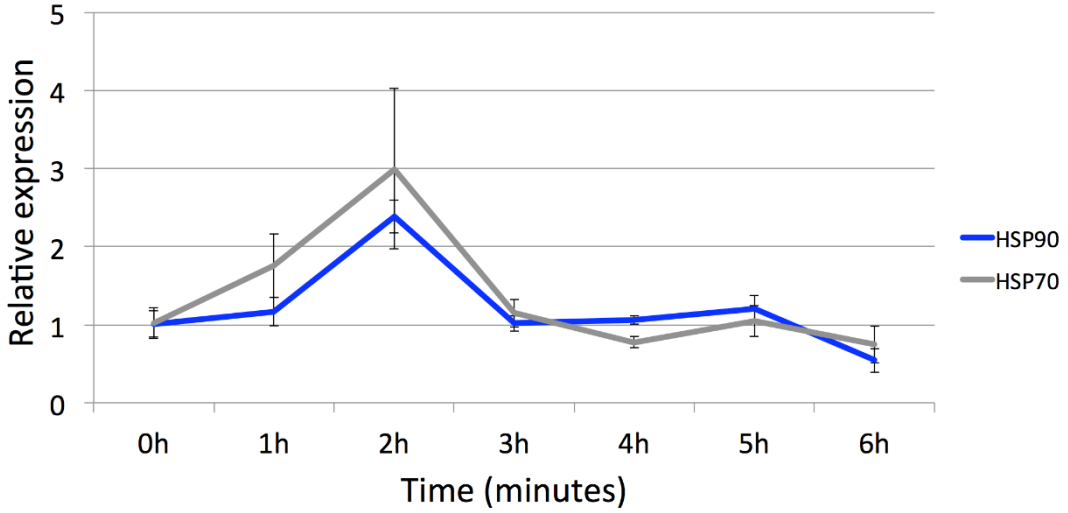
Fresh Water Effects on Adult Lice – Osmotic Stress

Heat Shock Proteins (HSP):

- Stressful conditions ➔ cell response ➔ the upregulation of the HSPs
- many HSPs perform chaperone function - help to refold proteins that were damaged by the cell stress



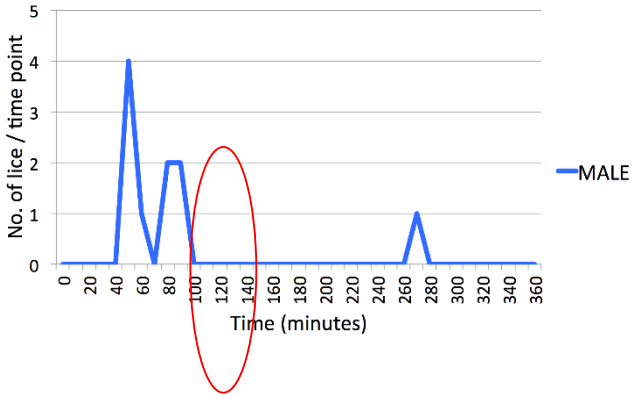
Adult male



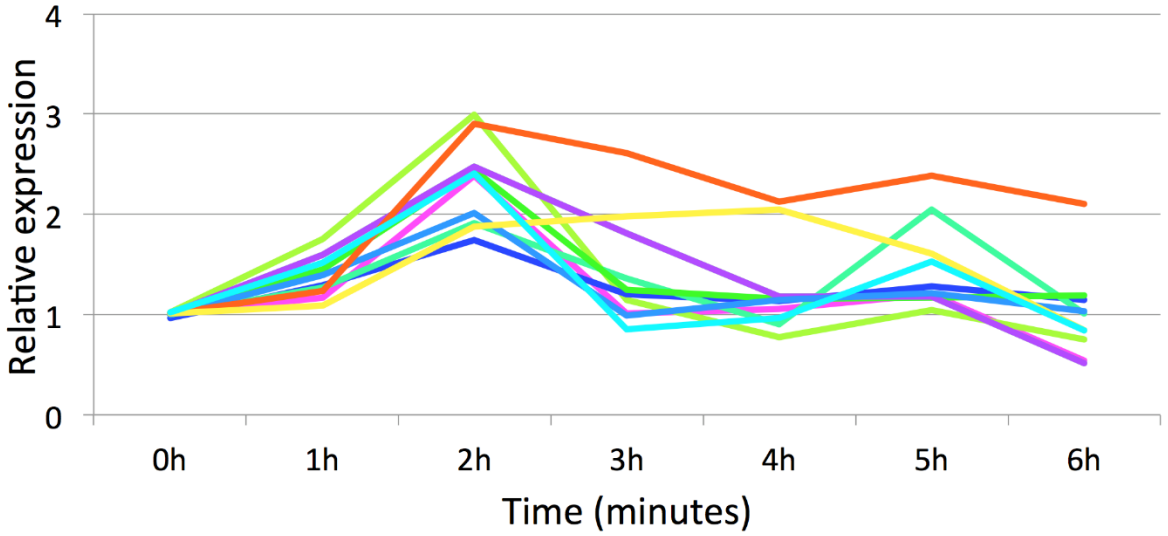
Fresh Water Effects on Adult Lice – Ion Exchangers/Transporters

Ion Exchangers/Transporters

- pump ions to/from cell with use od energy (ATP)
- involved in number of physiological processes including osmoregulation

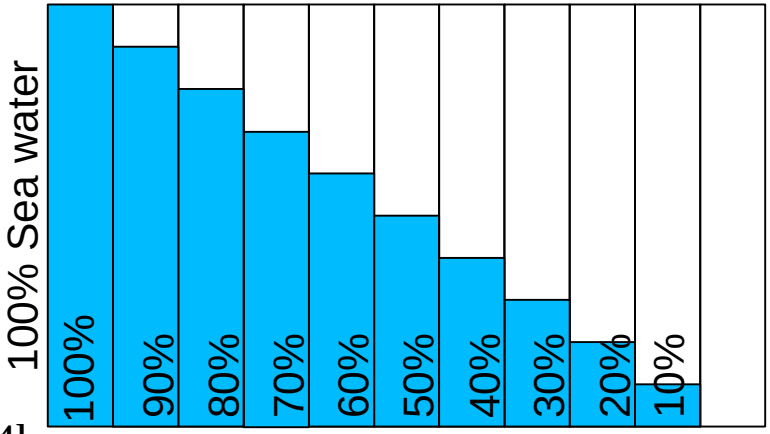


Adult male



Fresh Water Effects on Copepodids

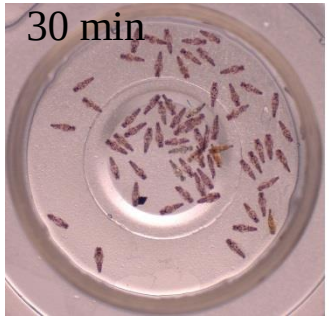
- Treatment time – 24h
- 11 groups – 100% – 0 sea water content.
- Evaluation: 30 min, 1h, 2h, 4h, 8h, 24h
  - Movement;
  - Staining;
- Result:
  - 100% - 40% sea water – 100% survival after 24h;
  - 30% - 20% sea water – 100% - 90% survival after 24h;

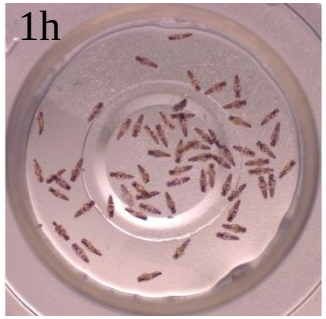


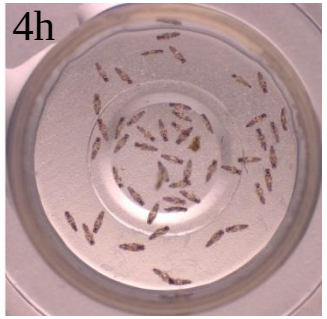
20% - 24h

CONTROL

  - 10%



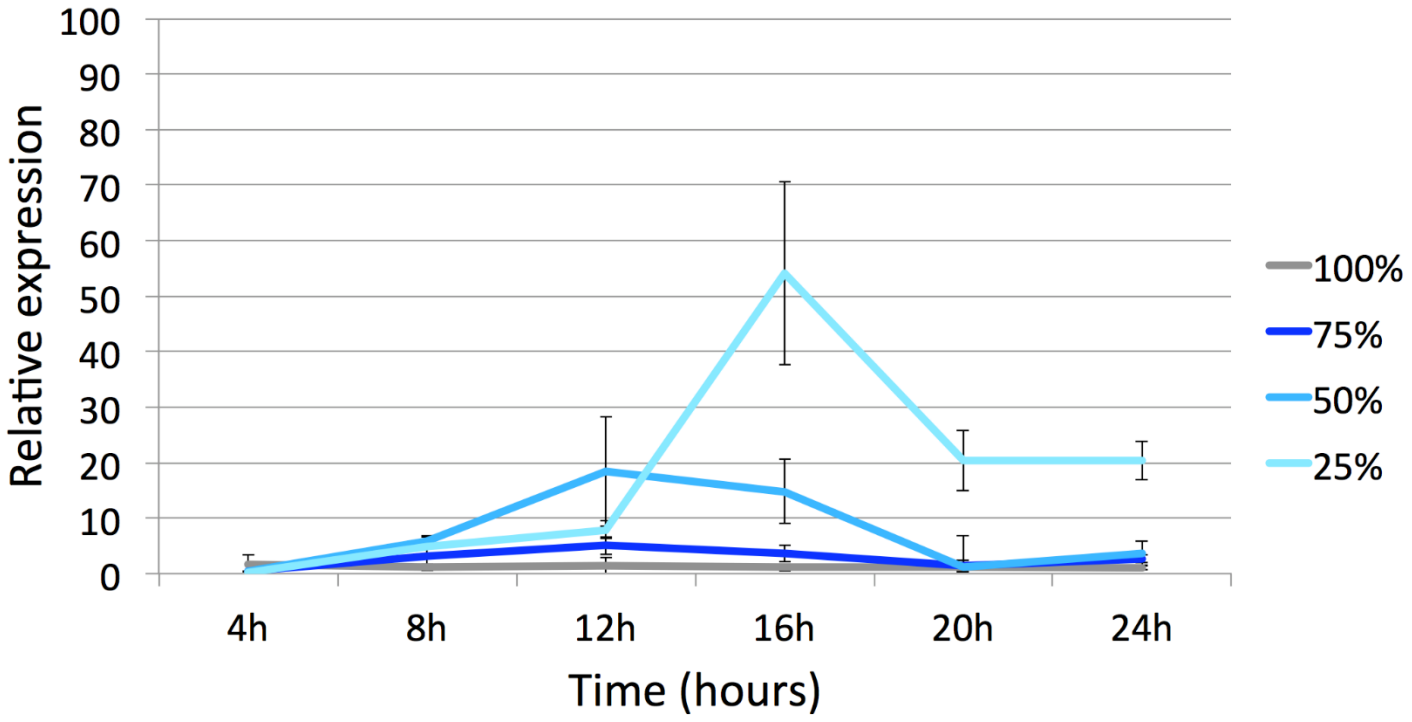




Fresh Water Effects on Copepodids – Osmotic Stress

Heat Shock Proteins 70 (HSP70):

- Copepodids – 100%, 75%, 50% and 25% sea water



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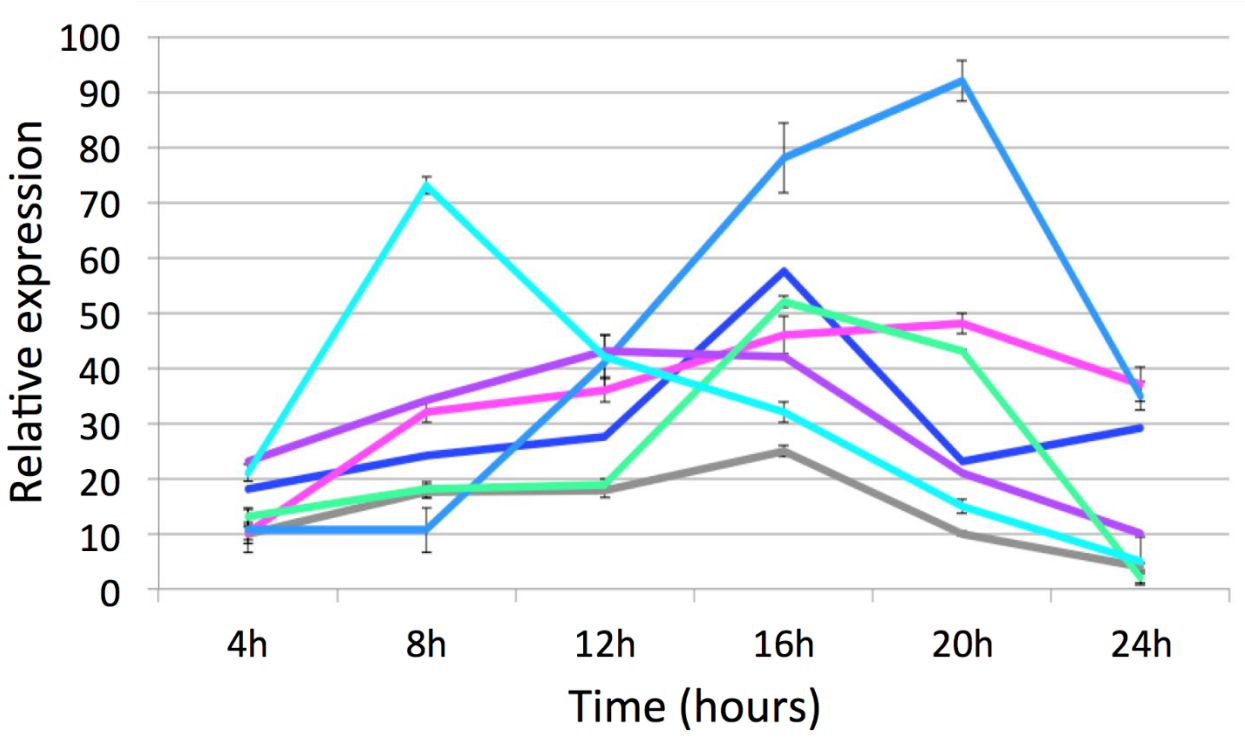
Fresh Water Effects on Copepodids – Ion Exchangers/Transporters

Ion Exchangers/Transporters:

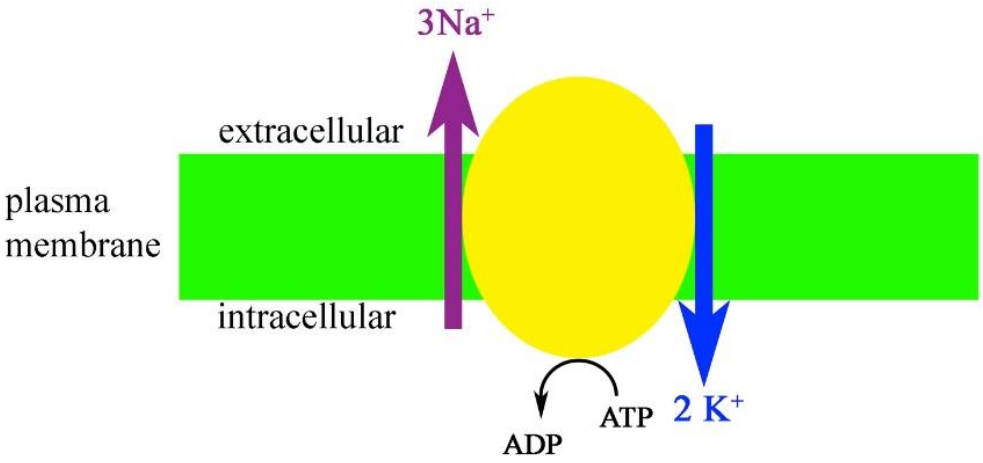
- Copepodids – 25% sea water



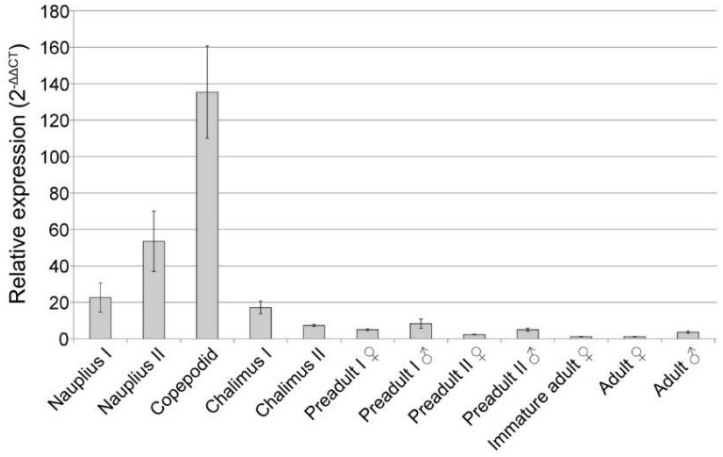
LsalNa^+/K^+-ATPase – Osmoregulation

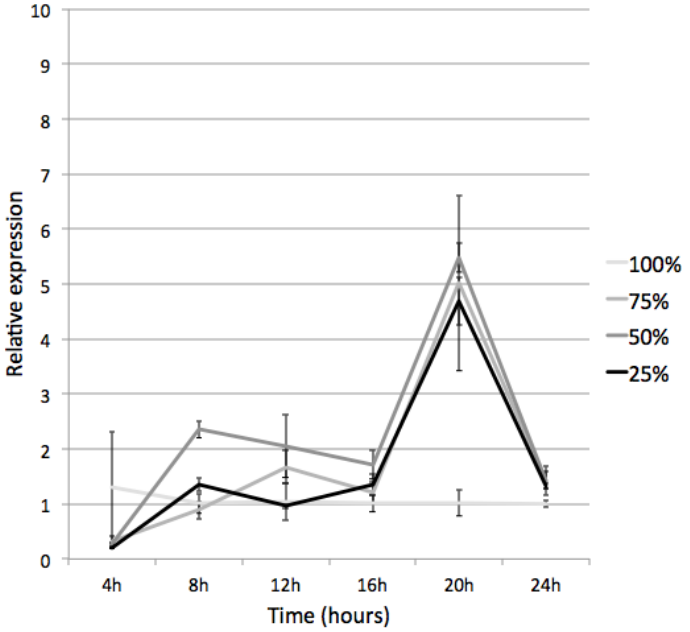
Sodium-Potassium ATPase

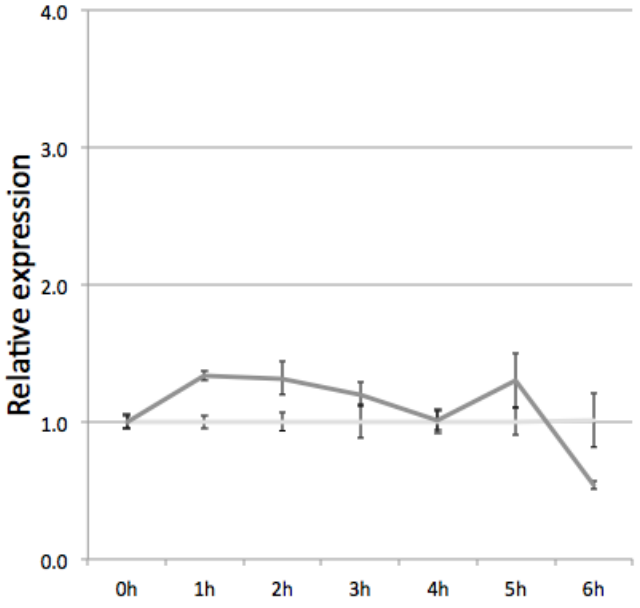
- Na^+ ions out of the cell and K^+ to the cell;
  - osmoregulation
  - cell volume regulation;
  - membrane excitability;
  - transport of nutrients;



*Lsal*Na$^+$/K$^+$-ATPase – Osmoregulation

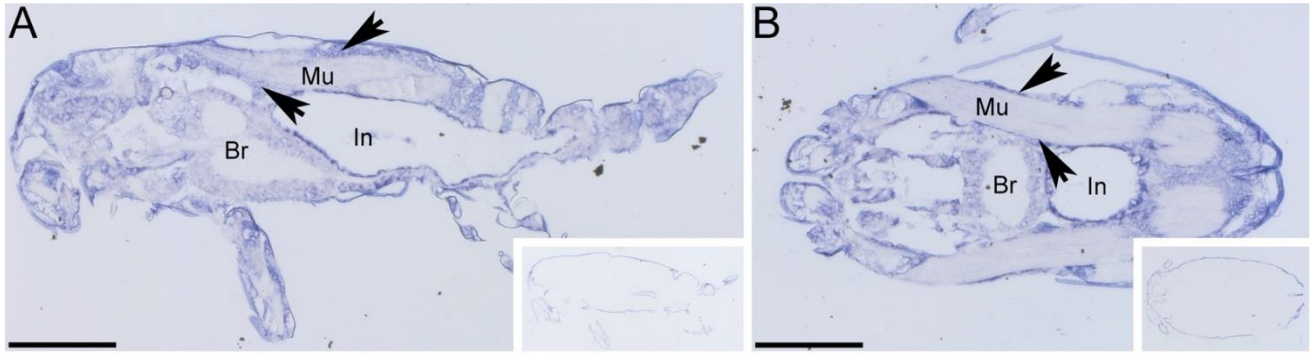




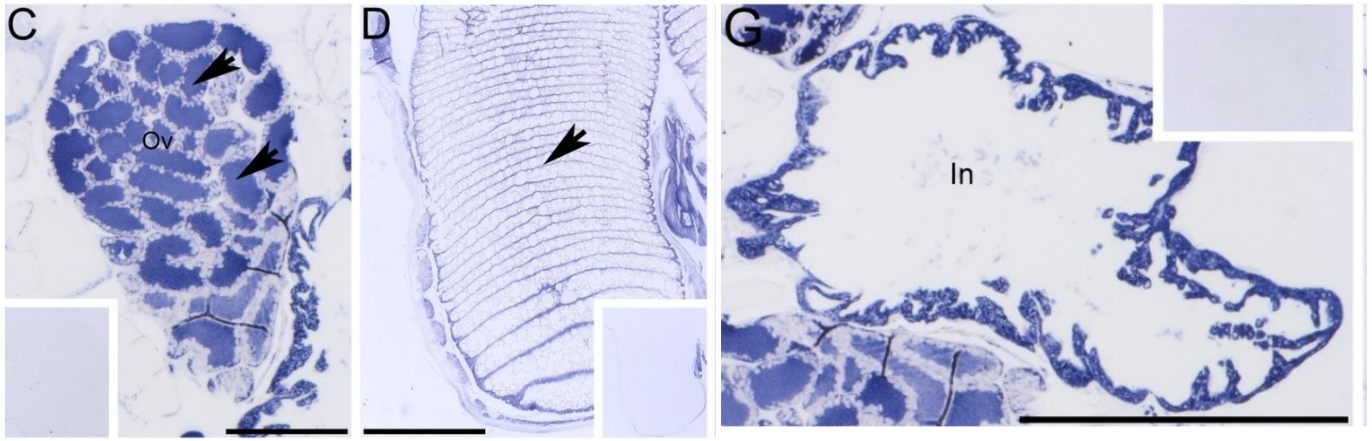


*Lsal*Na$^+$/K$^+$-ATPase – Gene Expression

copepodid



adults



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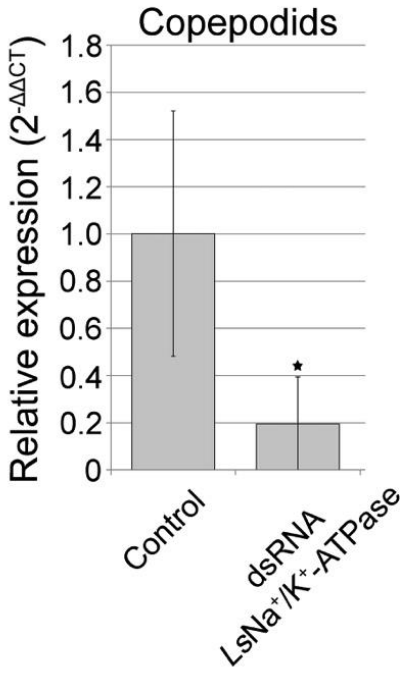
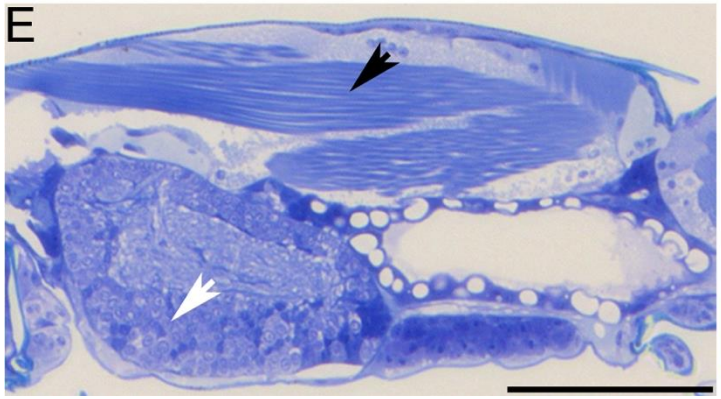
*Lsal*Na$^+$/K$^+$-ATPase – RNAi Copepodids

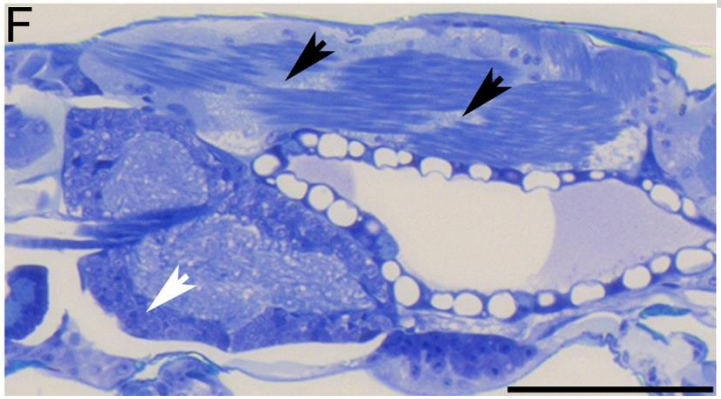
Movement impairment:

➔ Water/Ions transport

➔ Defects in muscle development







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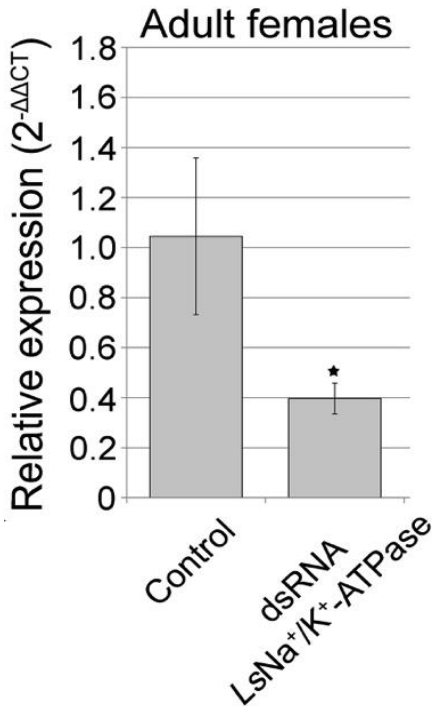
*Lsal*Na$^+$/K$^+$-ATPase – RNAi Adult Females



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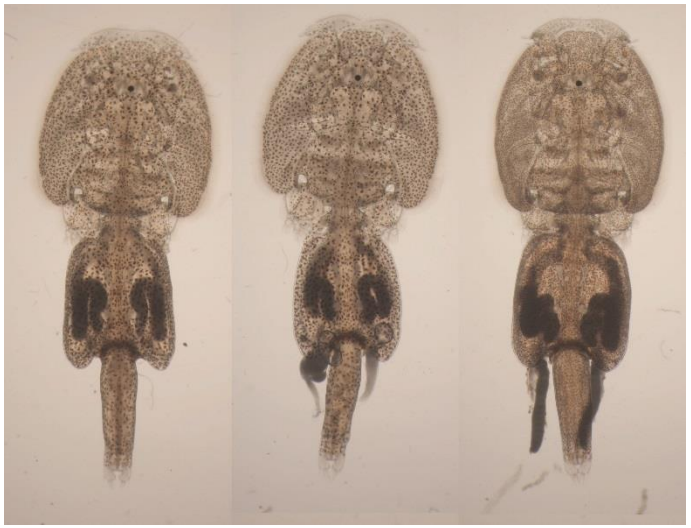

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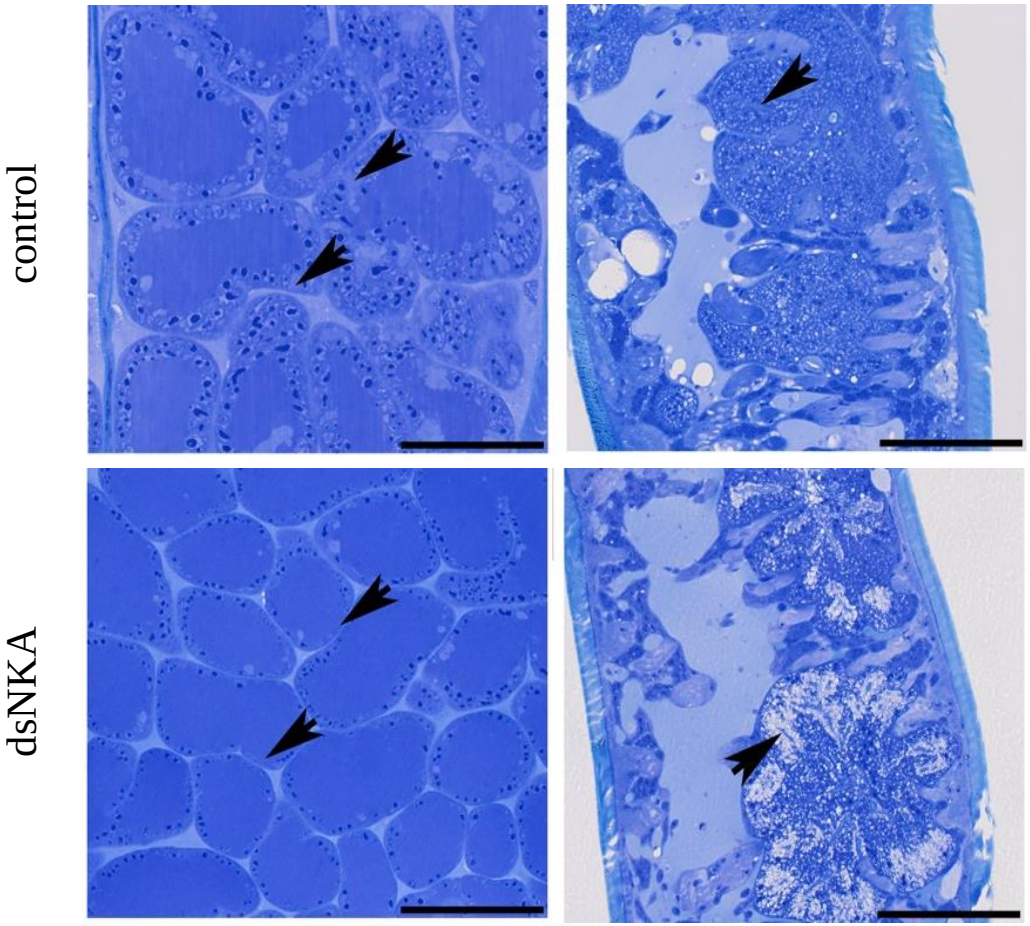
LsalNa^+/K^+-ATPase – RNAi Adult Females

- Defects in reproductions
- Defects in glands



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OSMO_LUS – FHF project: 901208

Acknowledgements

- Frank Nilsen
- Heidi Kongshaug
- Per Gunnar Espedal
- Lars Hamre