

**Do Short-Chain Chlorinated Paraffins Have Endocrine
Disrupting Potential?
Molecular And Demographics-Based Investigation In The
Rotifer *Brachionus calyciflorus***

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Take home message:

SCCP = endocrine disruption in *B. calyciflorus*

Concentrations $\leq 1 \mu\text{g/L}$ = significant effects

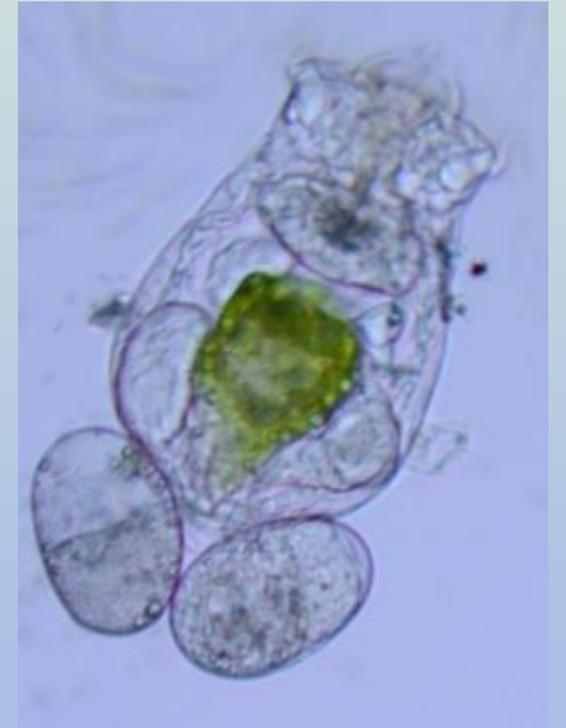
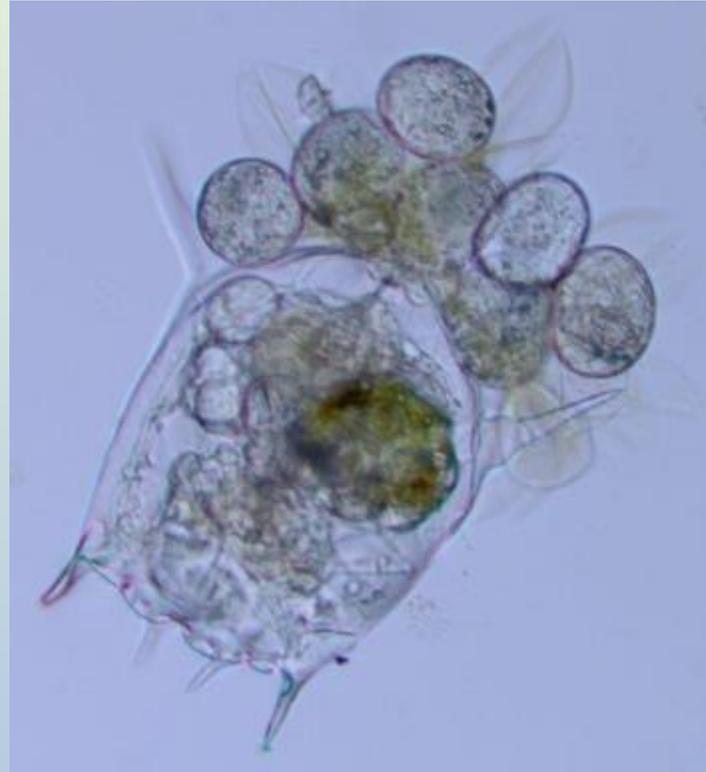
Possible population-level impacts?

Temperature-mediated effects

What is a rotifer?

- Zooplankton
- Filter feeder
- Body length < 0,5mm
- Fast pop. growth rate

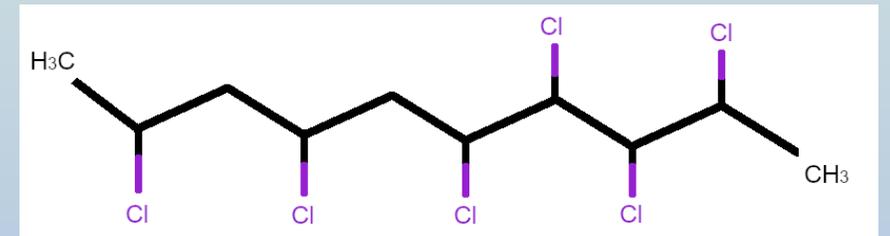
- Species: *Brachionus calyciflorus*
 - Useful laboratory model



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What about SCCPs?

- Linear alkanes (C₁₀-C₁₃), up to 70% chlorine mass
- Functions:
 - Industrial lubricants
 - Plasticizers
 - PVC processing
- Banned since 2018 (Stockholm Convention)
- Detected on all continents
 - In freshwaters: ng/L or µg/L range



10 carbon SCCP, 61% chlorine mass

SCCP characteristics

- Persistent – Bioaccumulable – Toxic = (PBT)
- Long range atmospheric transport
- Released from electronic waste

- In amphibians and freshwater fish
 - Developmental toxicity
 - Endocrine disruption

- On amphipods (*Gammarus pulex*): significant effects at 100ng/L

Problem & scope

- We know:
 - SCCPs toxic at environmental concentrations
 - Few data on invertebrates
 - Molecular biology = not always representative of whole animals
 - Effects can be t° -dependent

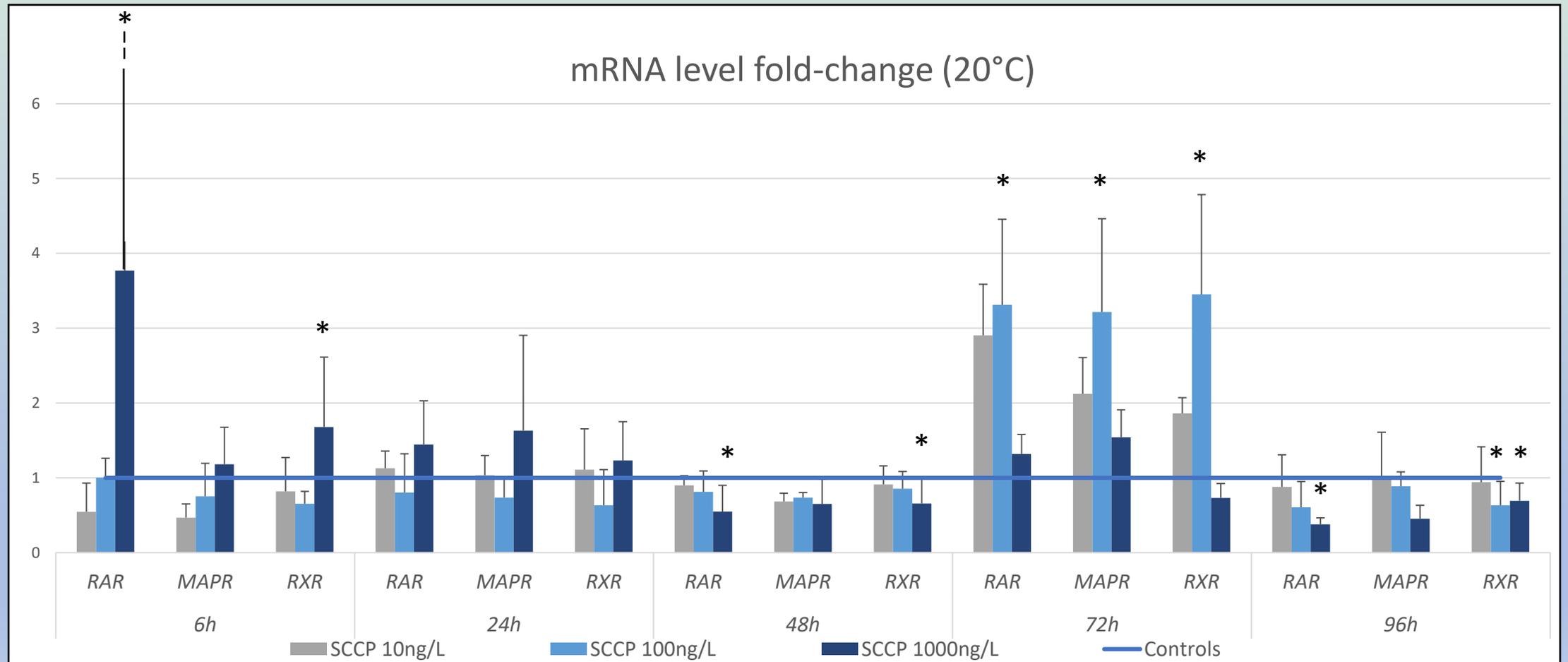
Can we combine assays at different levels of organisation to get a clearer picture of the real effect of contaminants and environmental conditions?

Methods:

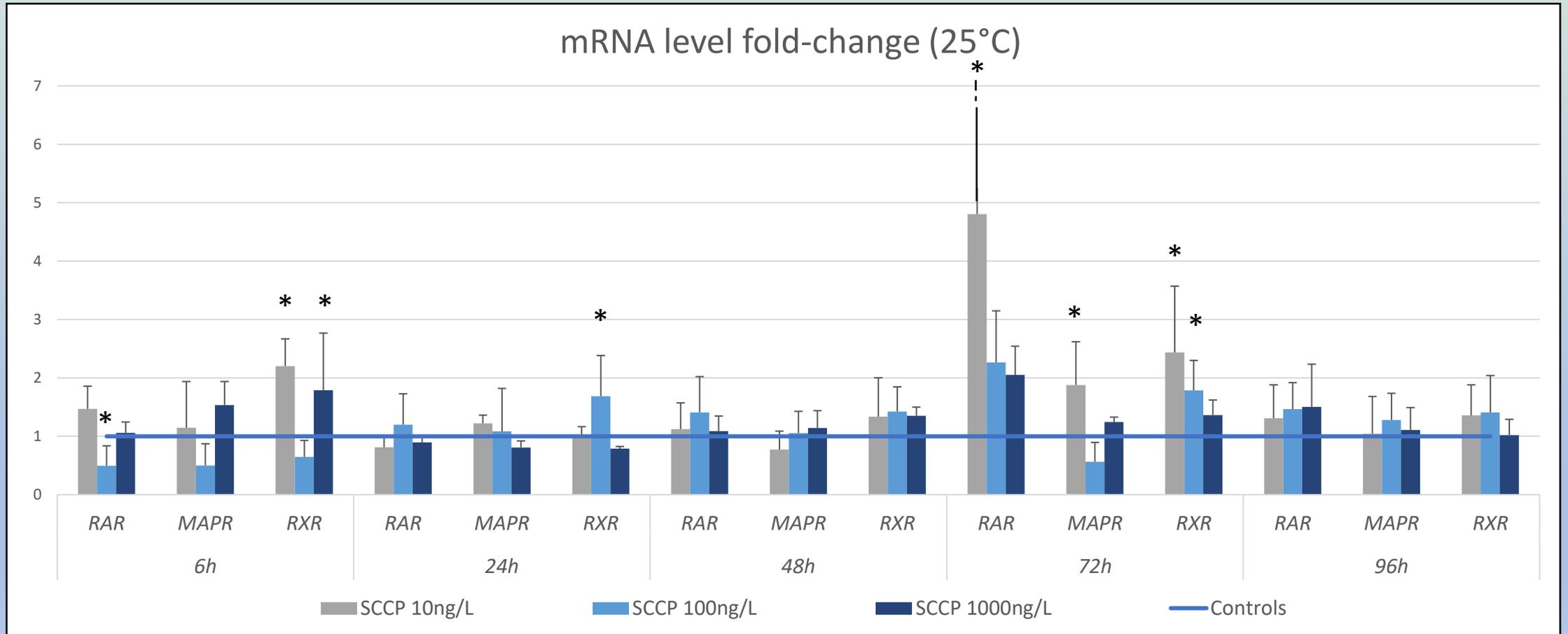
- 3 contaminant concentrations + controls
 - 10 – 100 – 1000ng/L in solvent (acetone)
- All assays realised at two t°
 - 20 or 25°C, with 4°C daily fluctuation of temperature
- Duration:
 - Molecular biology: exposed 6 – 24 – 48 – 72 – 96 hours
 - Demographic parameters: counted at 48 – 72 – 96 hours

Studied genes	
Retinoid – A – receptor	<i>rar</i>
Membrane-associated progesterone receptor	<i>mapr</i>
Retinoid – X – receptor	<i>rxr</i>

Molecular biology:



Molecular biology:

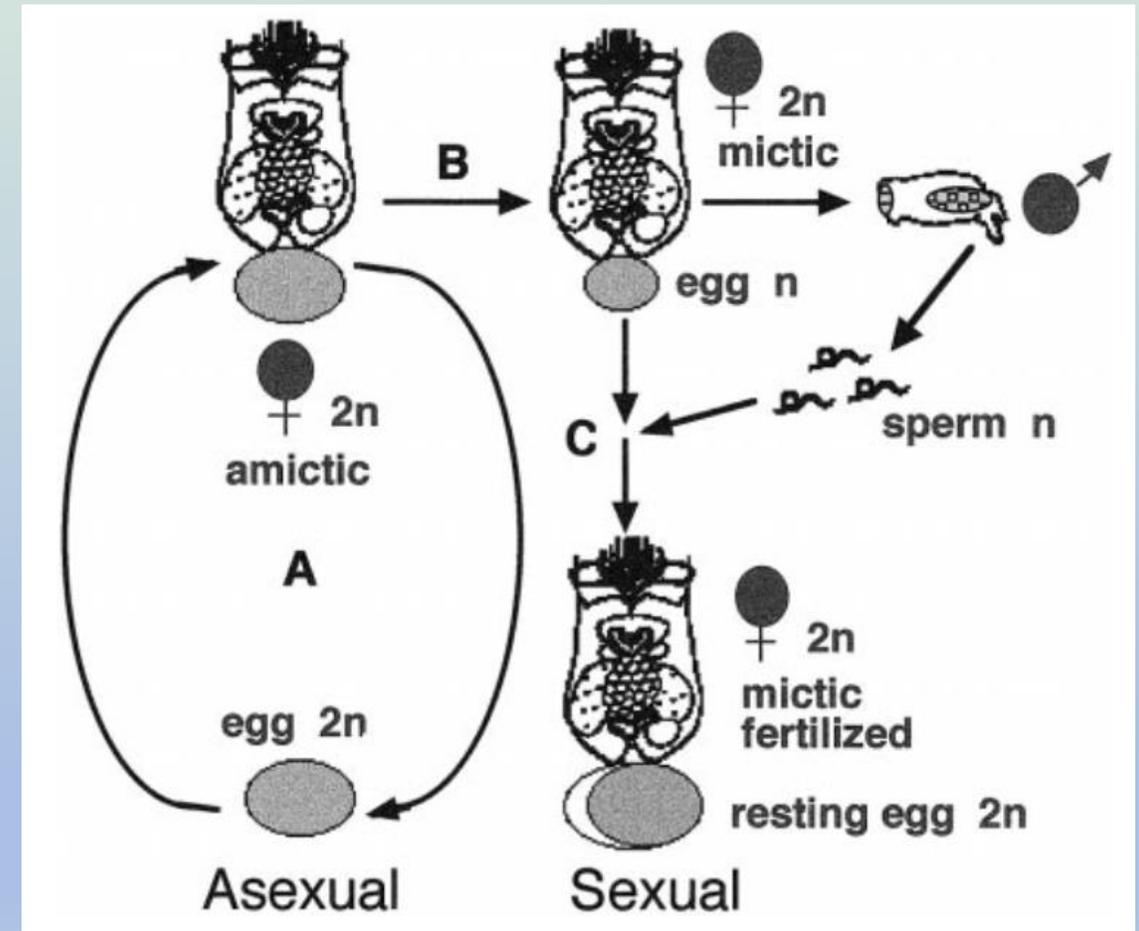


Discussion:

- Majority of effects: 6 & 72h
- 6h = initial toxicity then amictic female acclimatation?
- 72h = beginning of fertilization & resting egg production
- *rxr* & *rar* complementarity
- *mapr* and sexuality induction

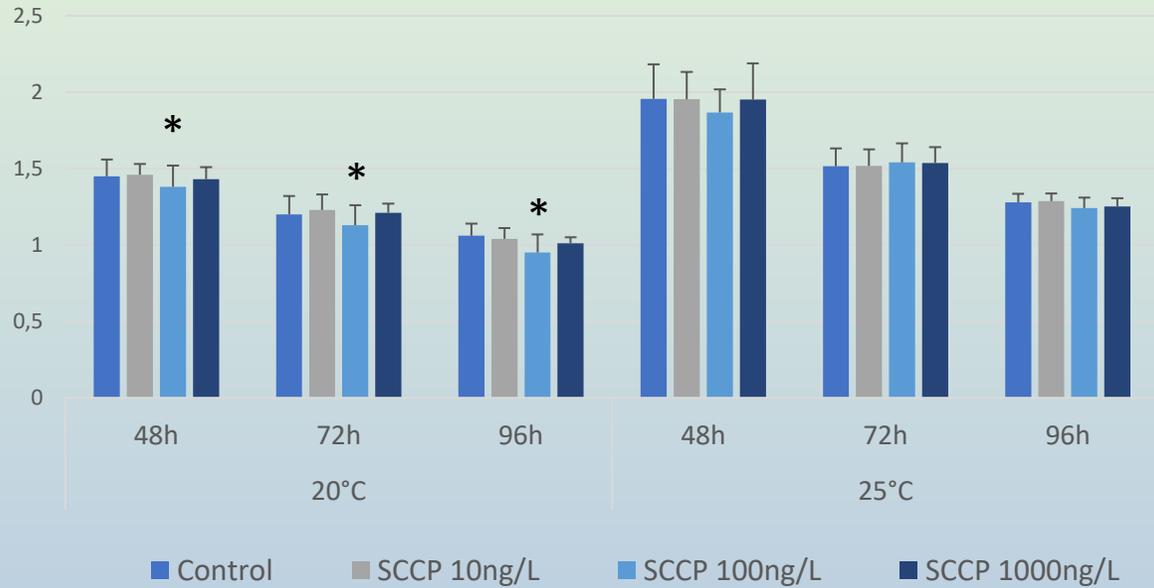
Demographic parameters:

- Intrinsic growth rate
 - Mixis rate
 - Fertilization rate
- Resting egg production
- Resting egg hatching

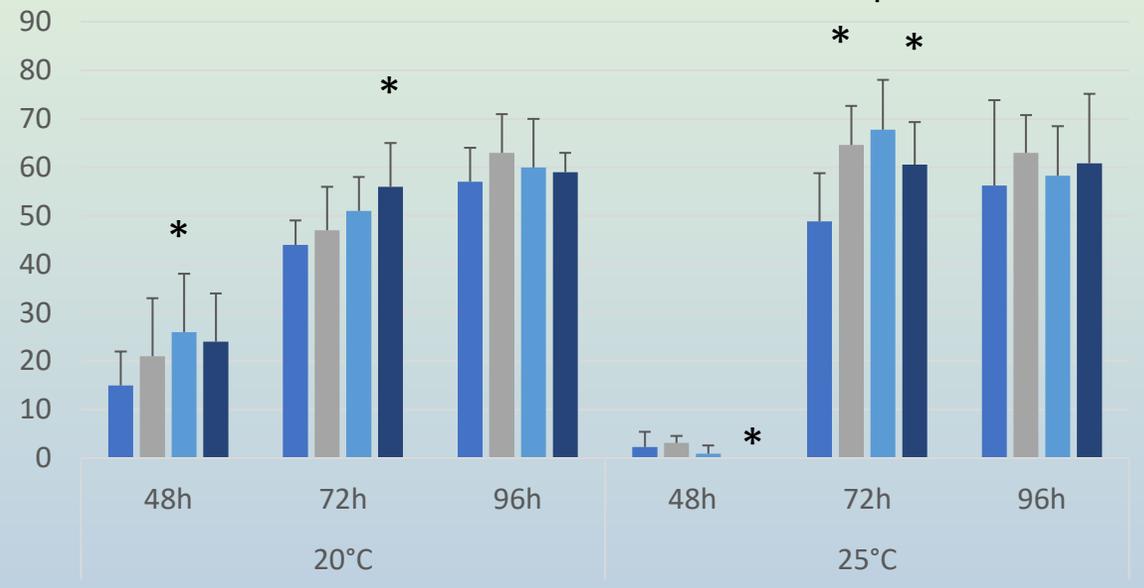


Preston, Snell, Robertson & Dingmann (2000). Use of freshwater rotifer *Brachionus calyciflorus* in screening assay for potential endocrine disruptors. *Environ. Toxicol. Chim.* Vol. 19-12. pp 2923 - 2928

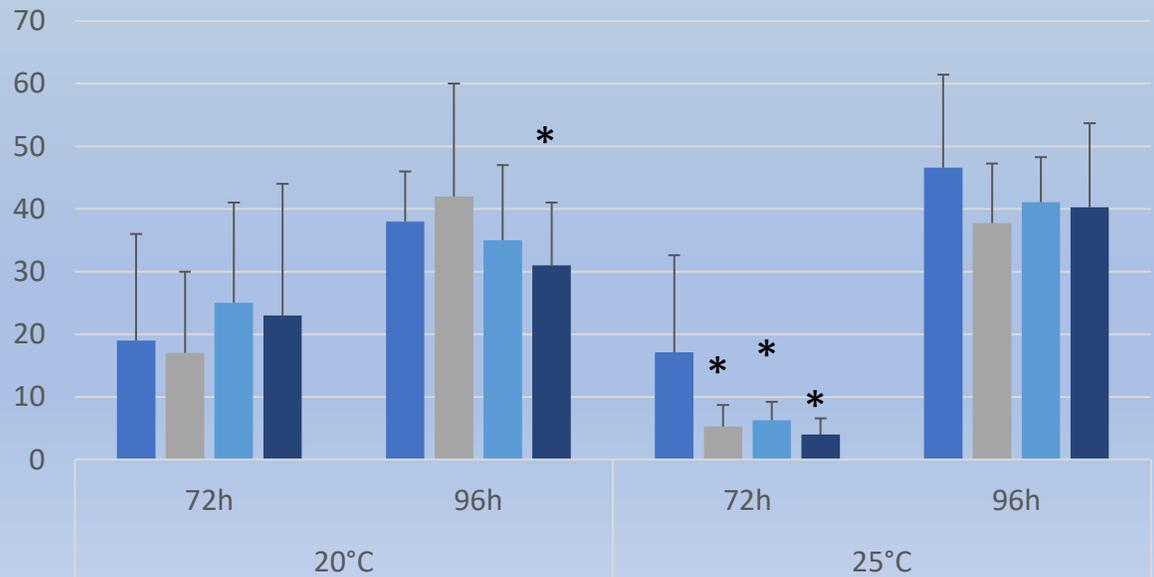
Intrinsic growth rate



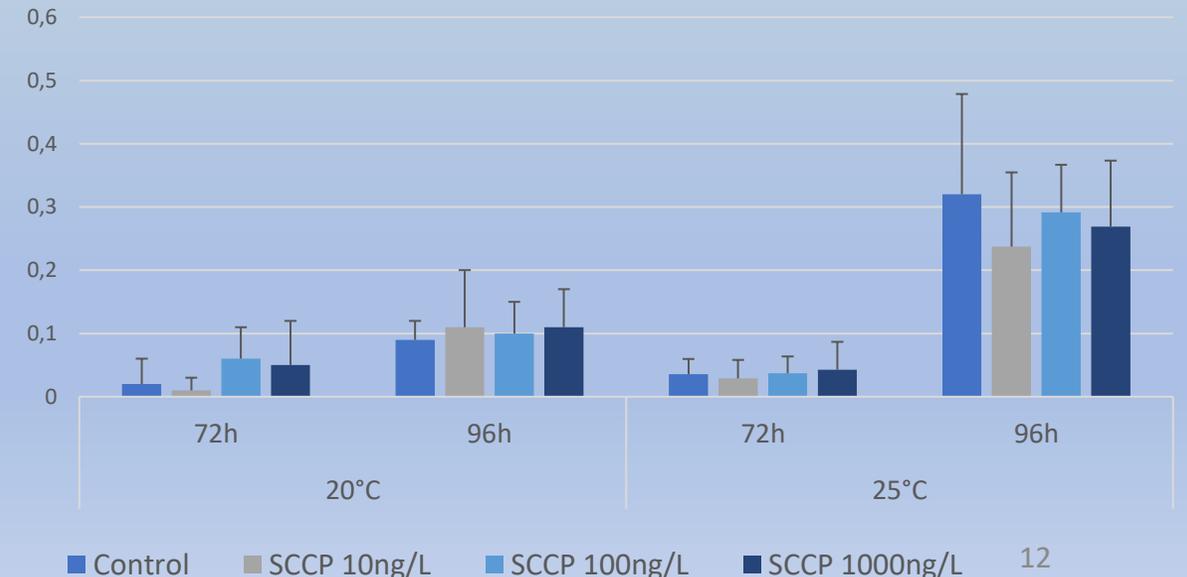
Mixis rate (%)



Fertilisation rate (%)



Resting eggs produced per female



Discussion:

- Stronger effects at 25°C
- On sexual reproduction:
 - Increase of mixis
 - Decrease of fertilisation
 - Resting egg production unaffected
- No remaining significant effects at 96h



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Conclusions:

- T° effect:
 - More affected mRNA levels at 20°C
 - More affected demographic parameters at 25°C
- Demographic effects seem short term, but multi-generational effects?
- Environmentally relevant SCCP concentrations affect *B. calyciflorus* endocrine receptors, mixis, and fertilisation

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