



**GHENT  
UNIVERSITY**

# Bio-actieve componenten uit algen getest met het gnotobiotisch Artemia systeem

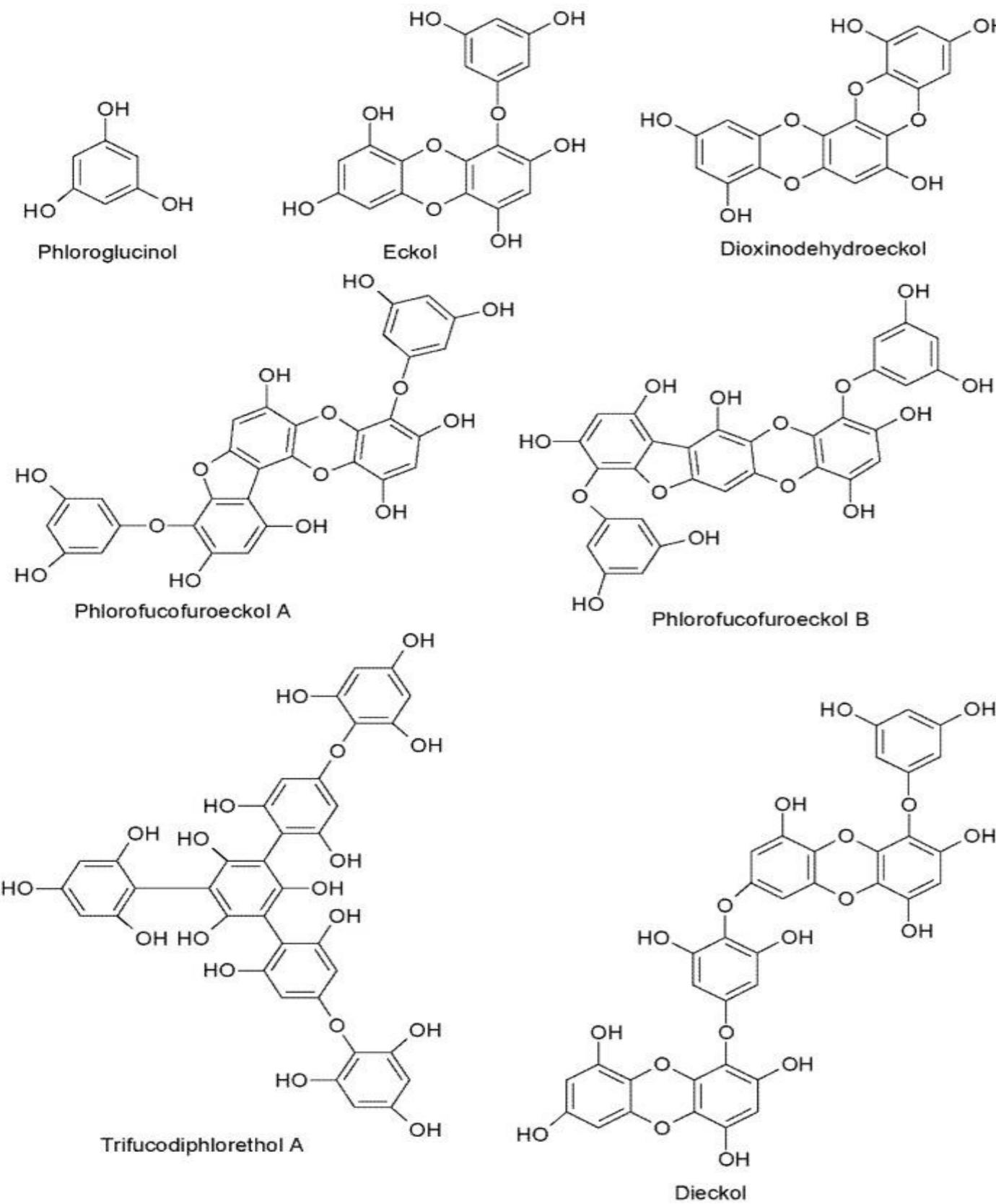
*Peter Bossier, Vikash Kumar, Kartik Baruah, Tania Lourenco*

*Ghent University*

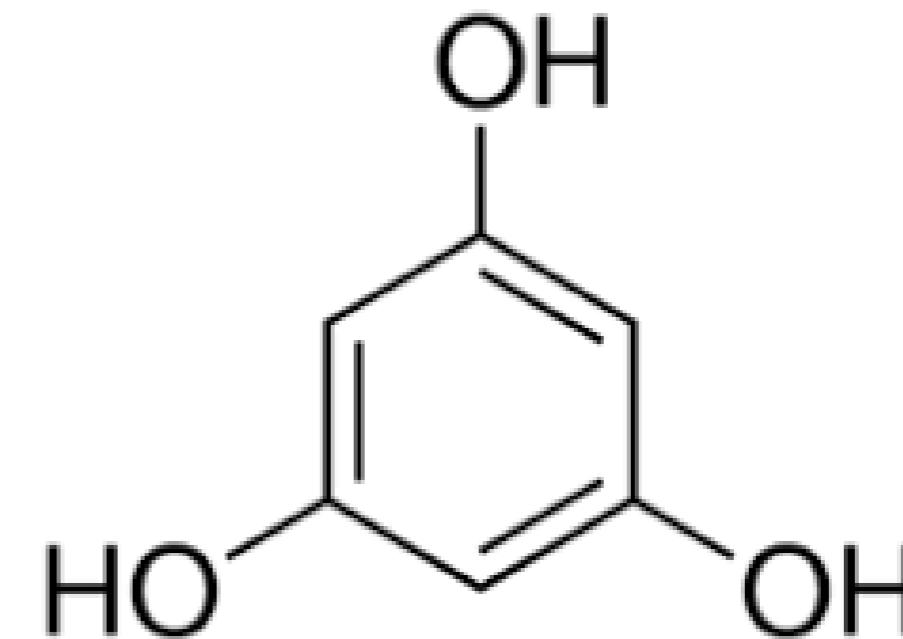
# MACROALGEN: PHLOROTANNINES

- Phlorotannines zijn een groep van complexe **polymeren van phloroglucinol** (1,3,5-trihydroxybenzene) uniek bij macroalgen.
- Deze fenolische componenten zijn
  - integrale structurele componenten van de celwanden van bruinwieren,
  - belangrijke secondaire ecologische rol in
    - Bescherming tegen UV straling
    - Bescherming tegen begrazing
  - In vitro: “anti-diabetic, anti-cancer, anti-oxidation, antibacterial, radioprotective en anti-HIV eigenschappen”

# PHLOROTANNINS

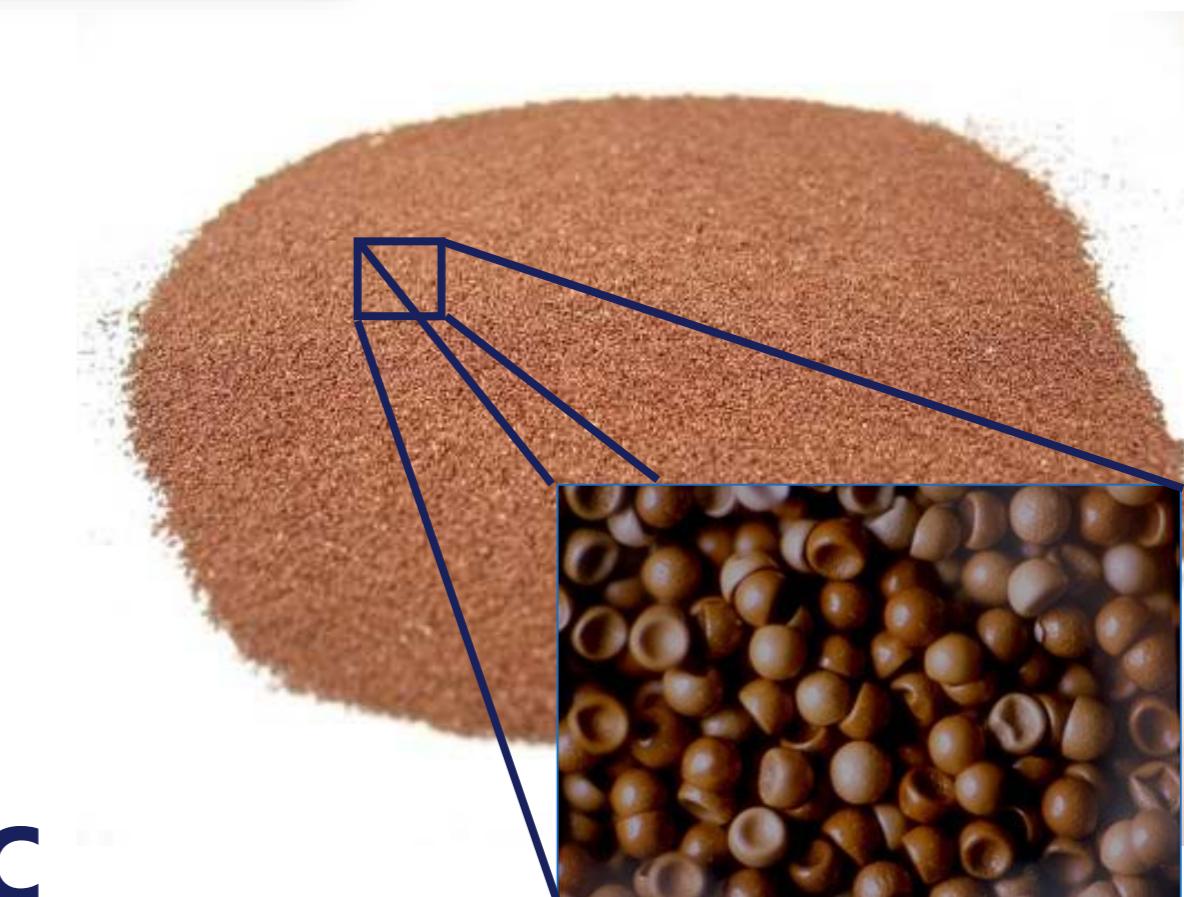
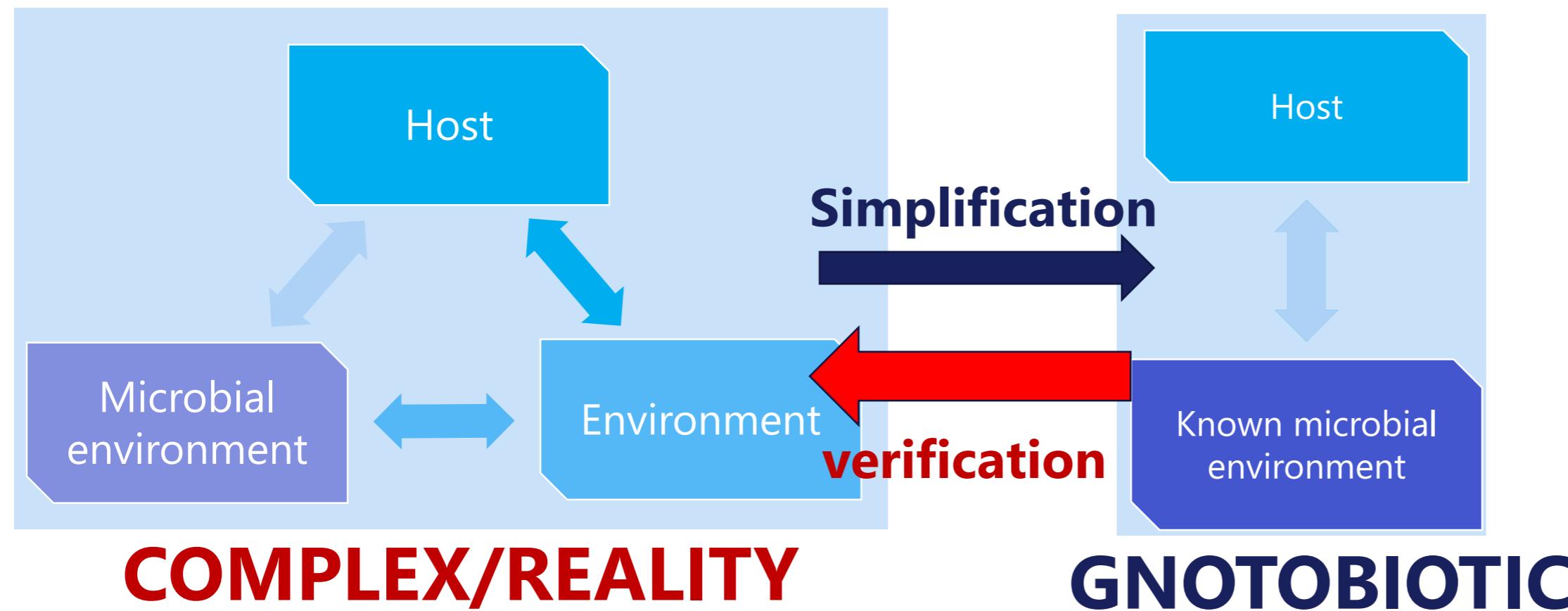
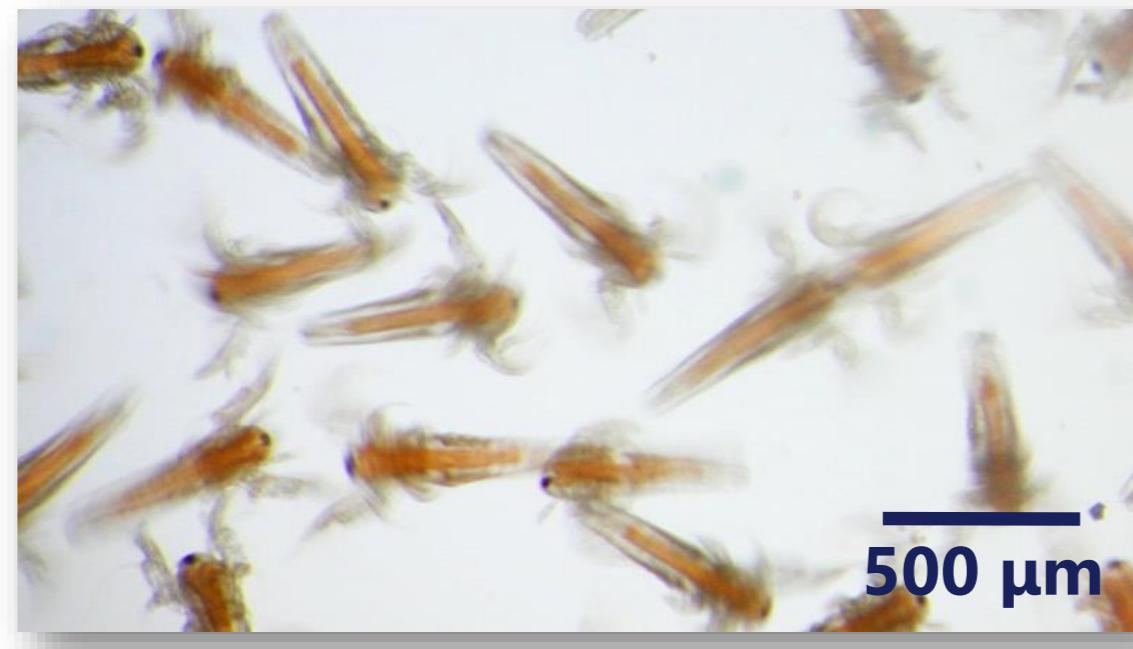


# PHLOROGLUCINOL



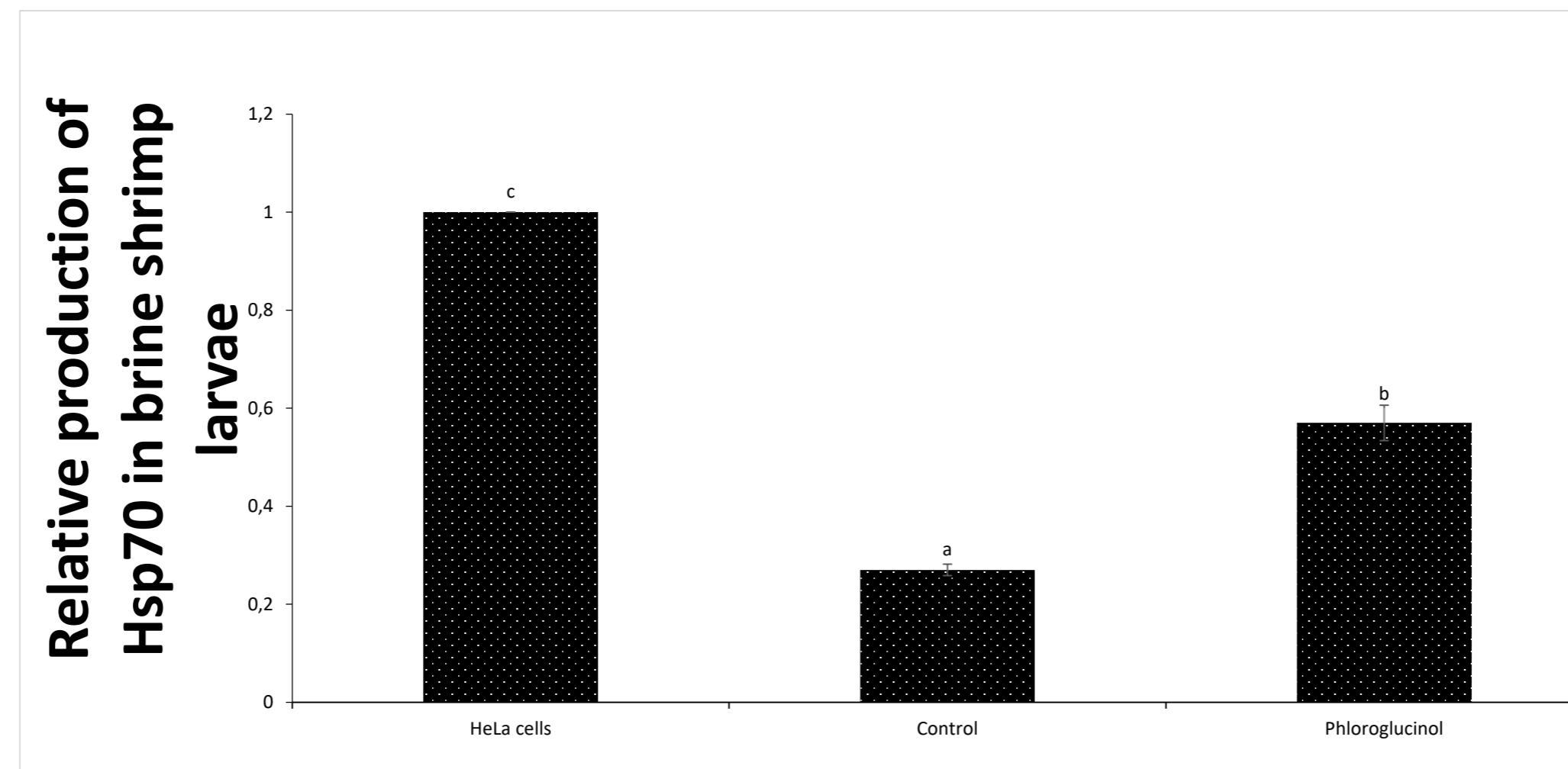
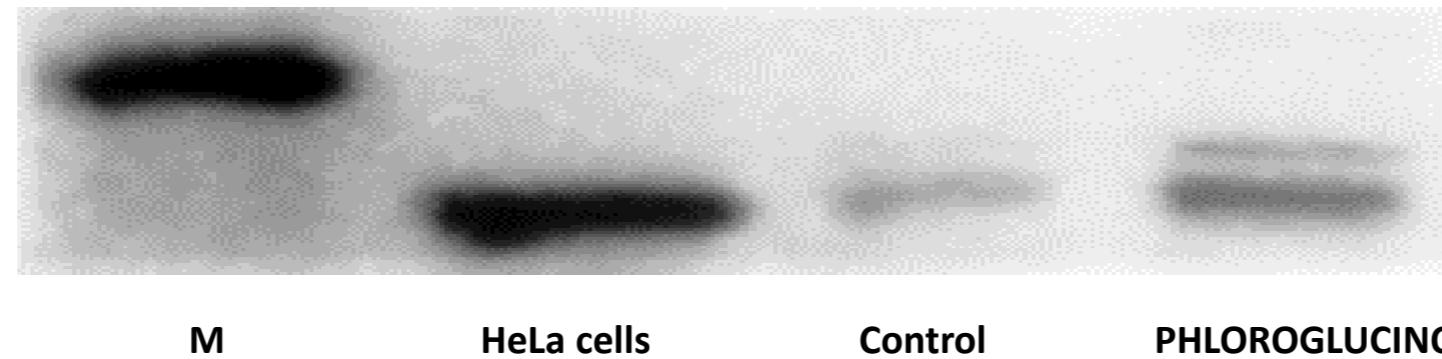
# Gnotobiotic *Artemia* screening platform

- Small scale (ml)
- Continuous supply
- High throughput (48-72h)
- Highly controllable
- Realistic response

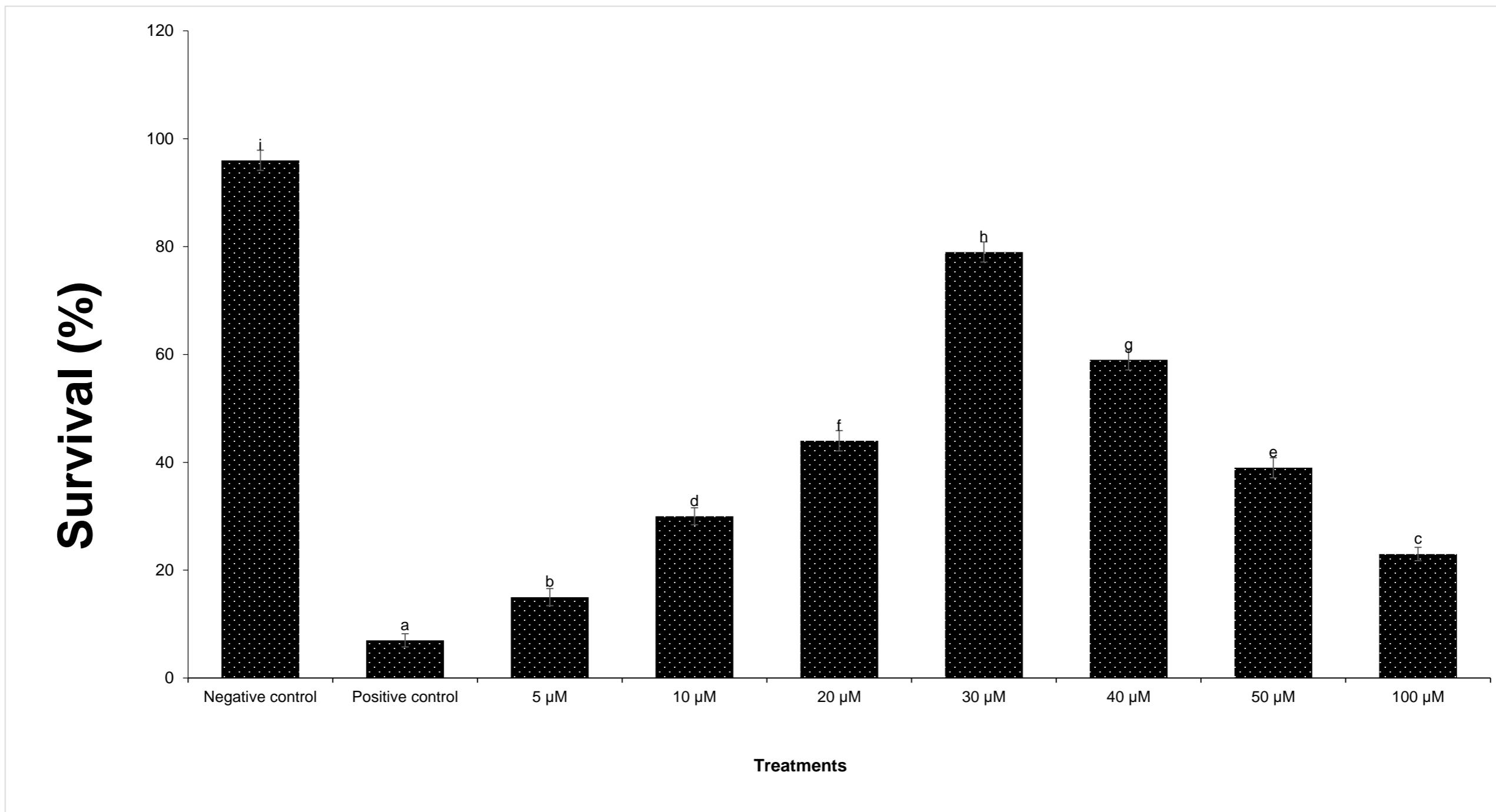


# PHLOROGLUCINOL ACTIVITY

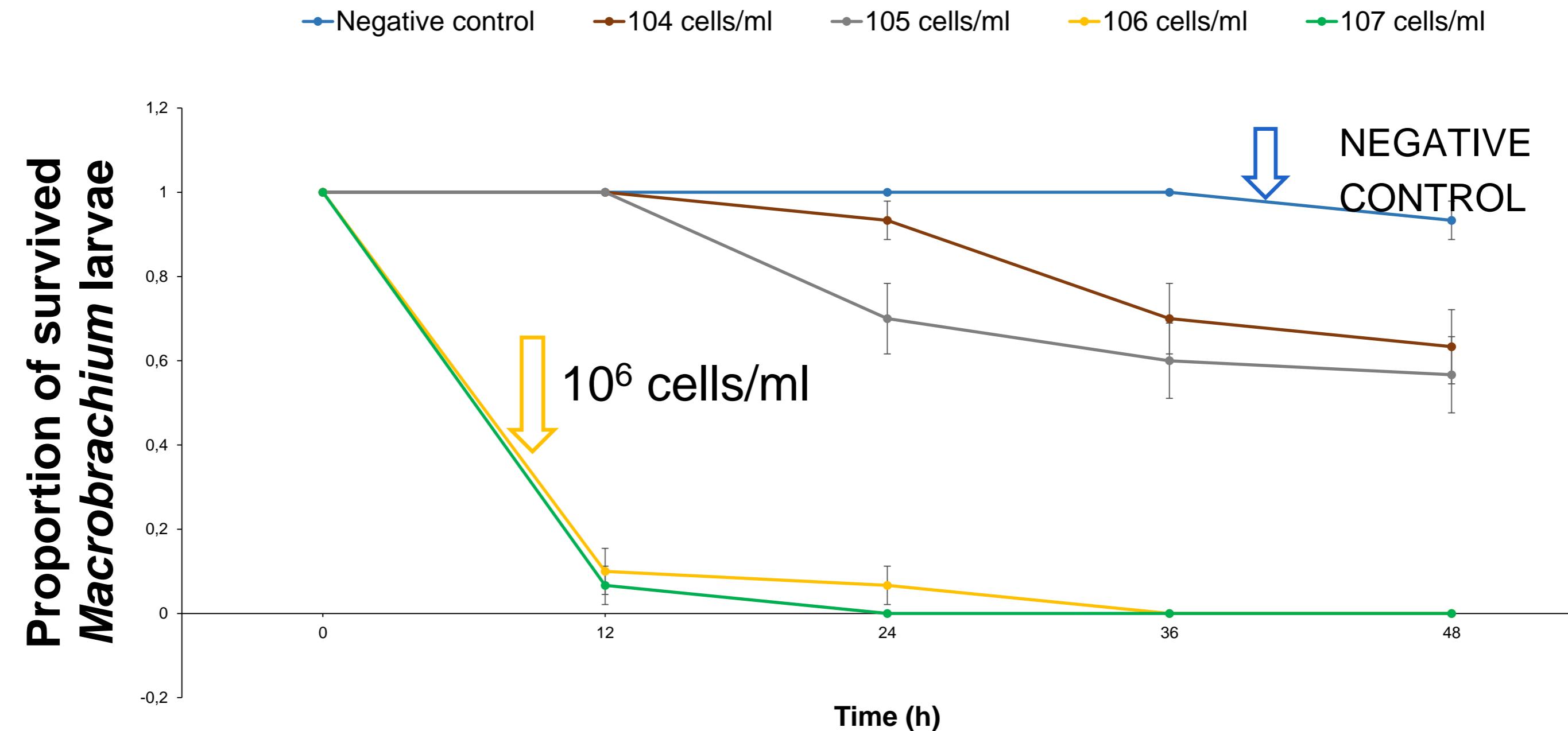
# INDUCTION OF HSP70 PROTEIN IN BRINE SHRIMP LARVAE PRETREATED WITH PHLOROGLUCINOL (30 µM).



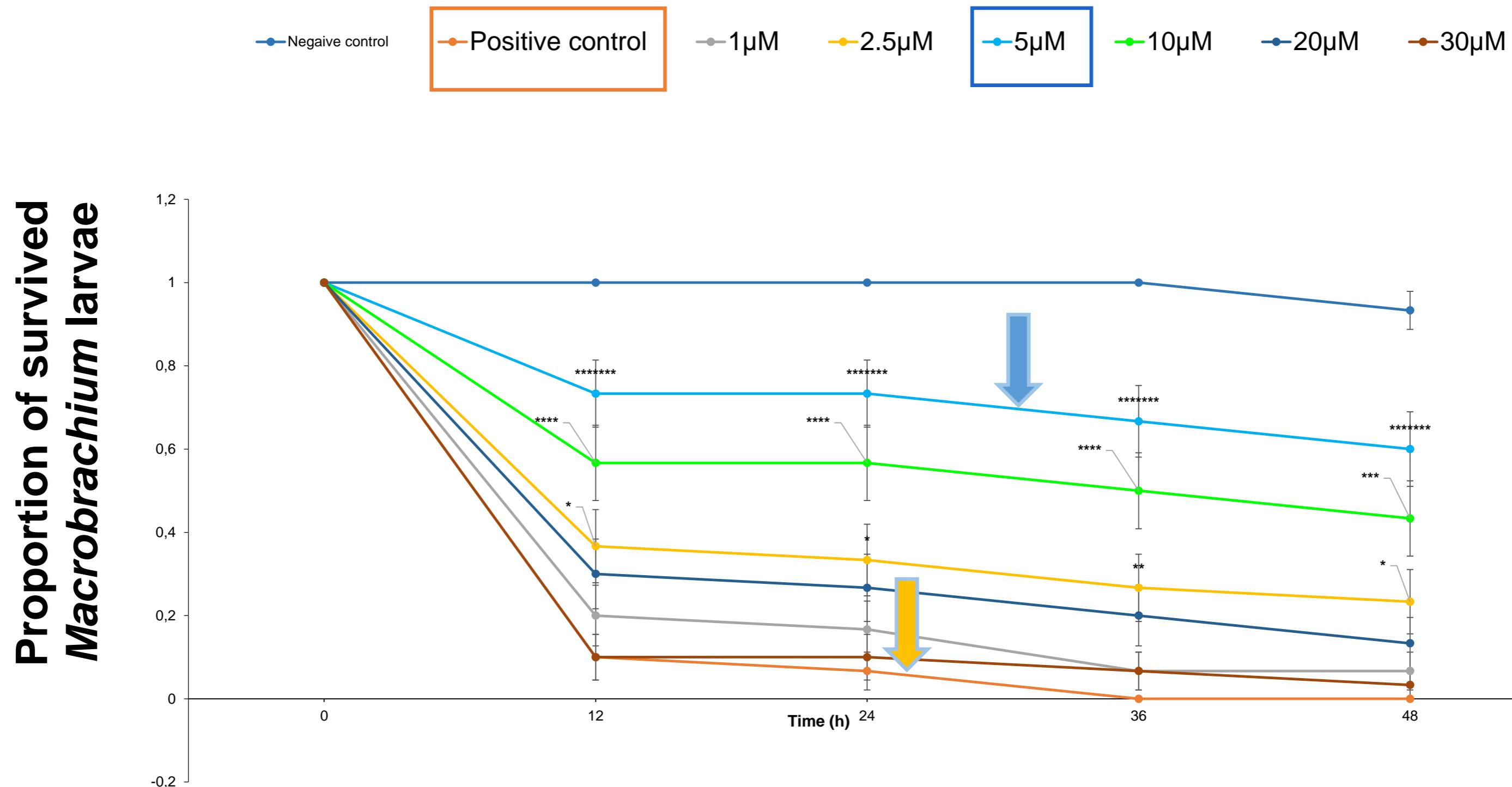
# PHLOROGLUCINOL CONFERS PROTECTION TO ARTEMIA AGAINST PATHOGENIC VIBRIO PARAHAEMOLYTICUS



# MACROBRACHIUM LARVAE (8-DAYS OLD) POST CHALLENGE WITH V. PARAHAEMOLYTICUS MO904 STRAIN



# EFFECT OF PHLOROGLUCINOL PRETREATMENT ON THE SURVIVAL OF MACROBRACHIUM LARVAE (8-DAYS OLD)



## CONCLUSION: PHOROGLUCINOL

- Non toxic at the tested concentration levels
- At 30 µM: induction of a stress response in Artemia
- Pretreatment with phloroglucinol confers protection against a biological stress (Vibrio)



## NEPTUNA

Novel Extraction Processes for multiple high-value compounds  
from selected Algal source materials

**Coordinator: Dr Dagmar Stengel, NUI Galway, Ireland**

Prof Bill Baker, NUI Galway and USF, US

Prof Alan Dobson, UCC, Ireland

Prof Peter Bossier, University of Ghent, Belgium

Dr Sarah Hoskins, Unilever, UK

Prof Jeannette Hammer Andersen, Norway

ERA-MBT 1<sup>st</sup> Transnational Joint Call: Biorefinery processes



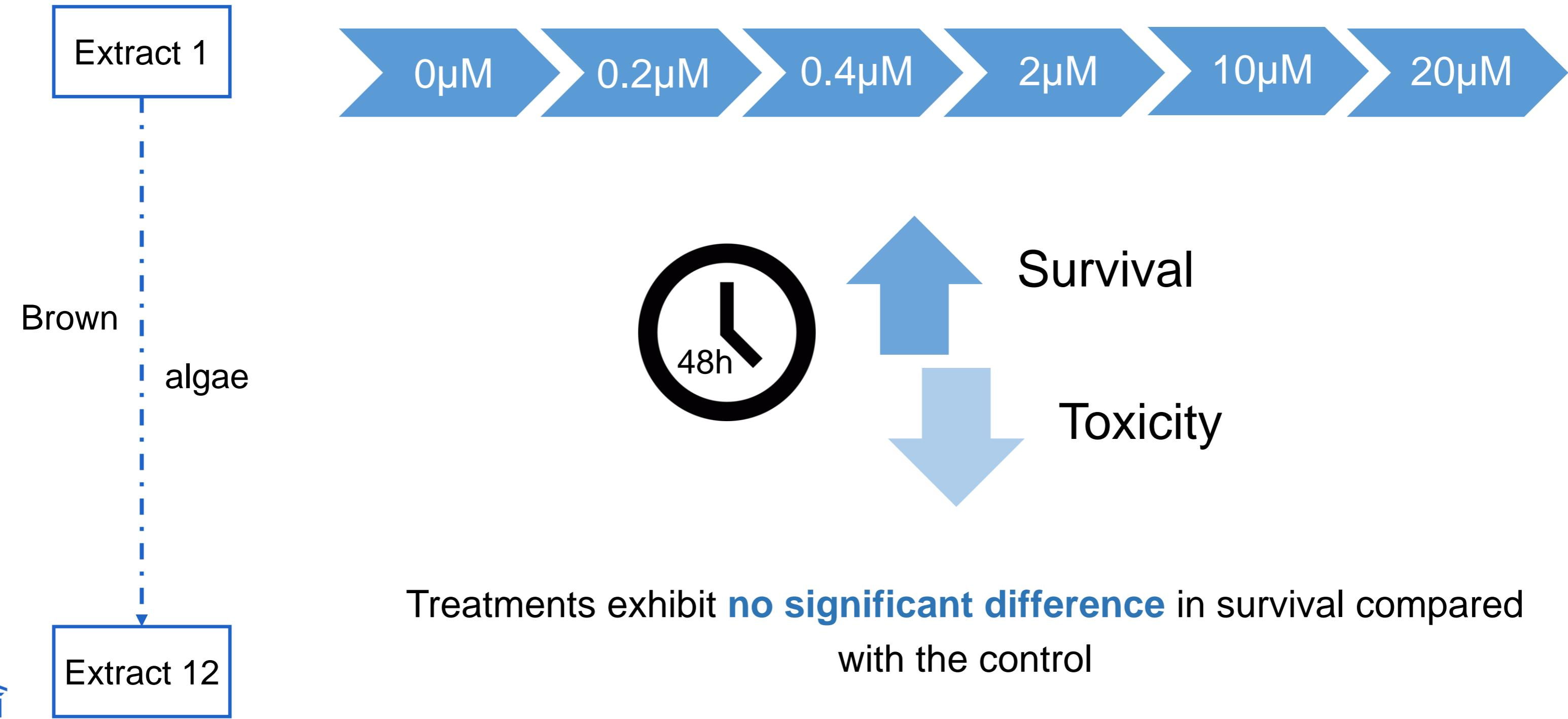
Marine Biotechnology ERA-NET (ERA-MBT) is funded under the European Commission's Seventh Framework Programme. Grant Agreement Number 604814 December 2013 - November 2017

# MACROALGAE EXTRACTS: PERFORMED TESTS

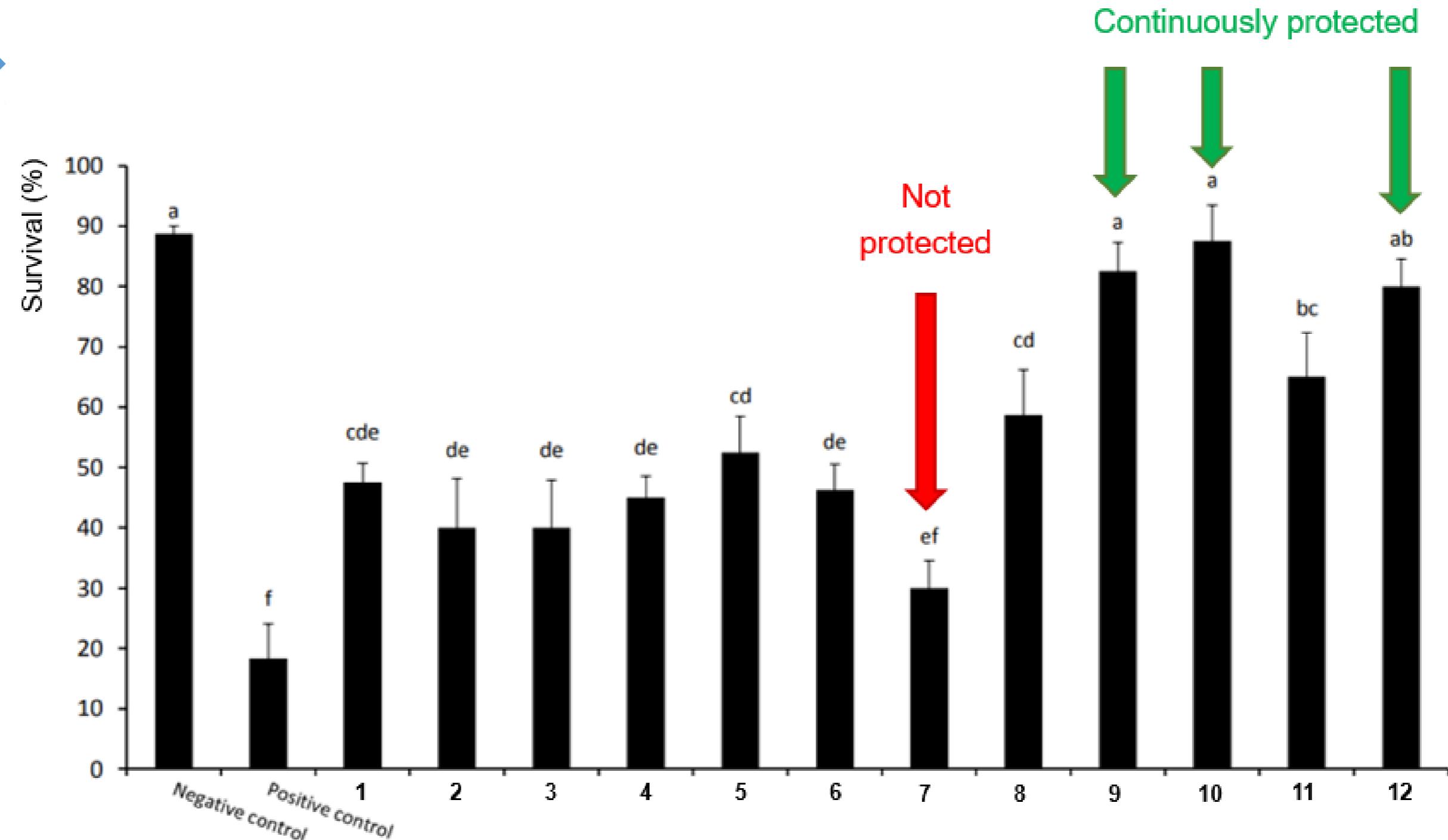
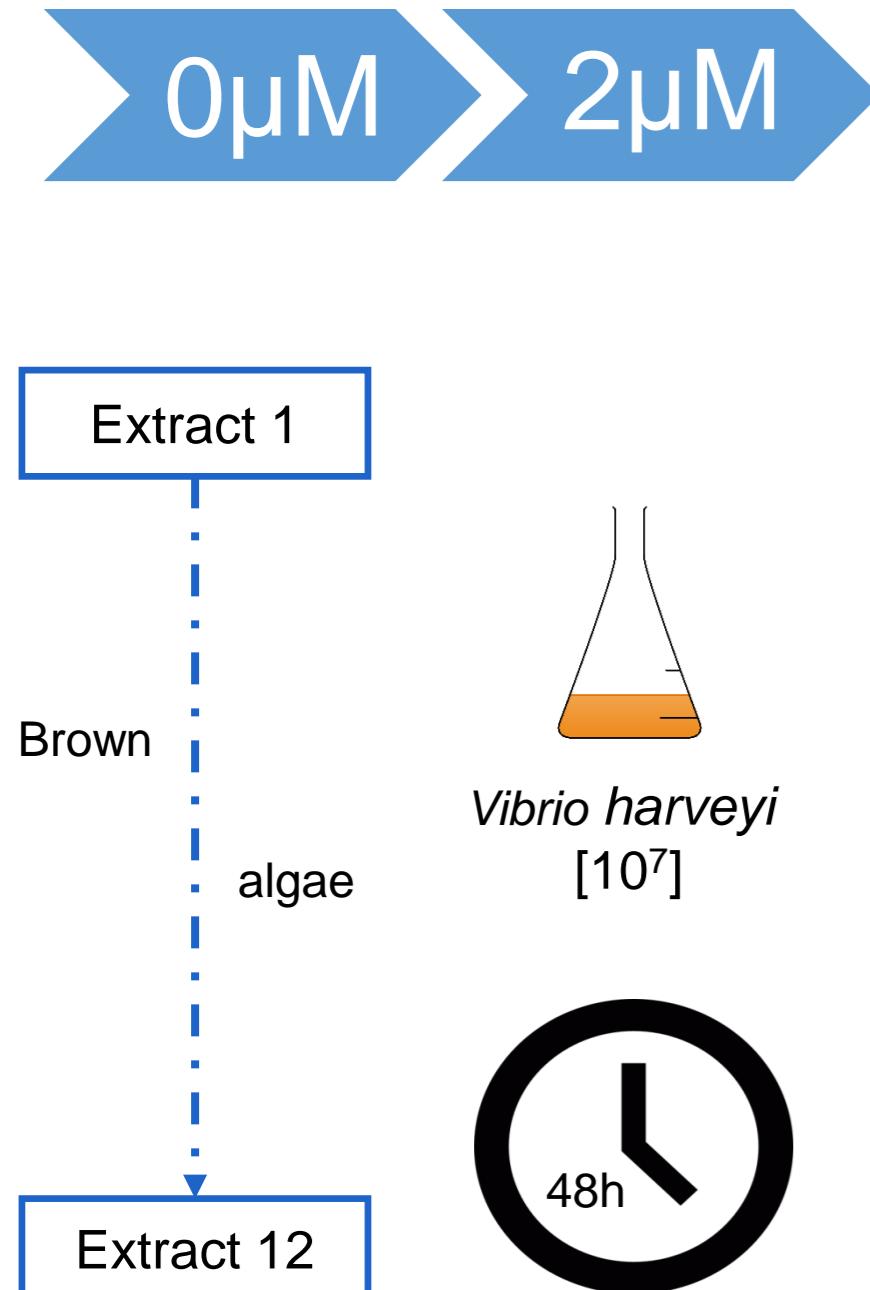
- ✓ In vivo toxicity testing algal extracts
  - ✓ Survival
- ✓ In vivo evaluation – anti-bacterial activity
  - ✓ Pre-treatment survival
  - ✓ Continuous treatment survival
- ✓ *V. harveyi* virulence phenotypes – effects
  - ✓ Growth
  - ✓ Luminescence

The total phenolic content (%) of the NEPTUNA extracts, was expressed as *phloroglucinol equivalent*

# IN VIVO TOXICITY – ALGAL EXTRACTS



# IN VIVO EVALUATION: ANTI-VIBRIO ACTIVITY



Effect of the algae extracts (AE) on the survival of Artemia larvae challenged with *V. harveyi*. Untreated Artemia challenged with *V. harveyi* (positive control) and those not challenged with *V. harveyi* (negative control) served as controls. All treatments were carried out with four replicates. Values (mean  $\pm$  standard error) indicated with different letters are significantly different ( $P < 0.05$ ).

# V. HARVEYI VIRULENCE PHENOTYPES – LUMINESCENCE

0 $\mu$ M > 2 $\mu$ M

Growth

No effect was seen on the growth of *V. harveyi*

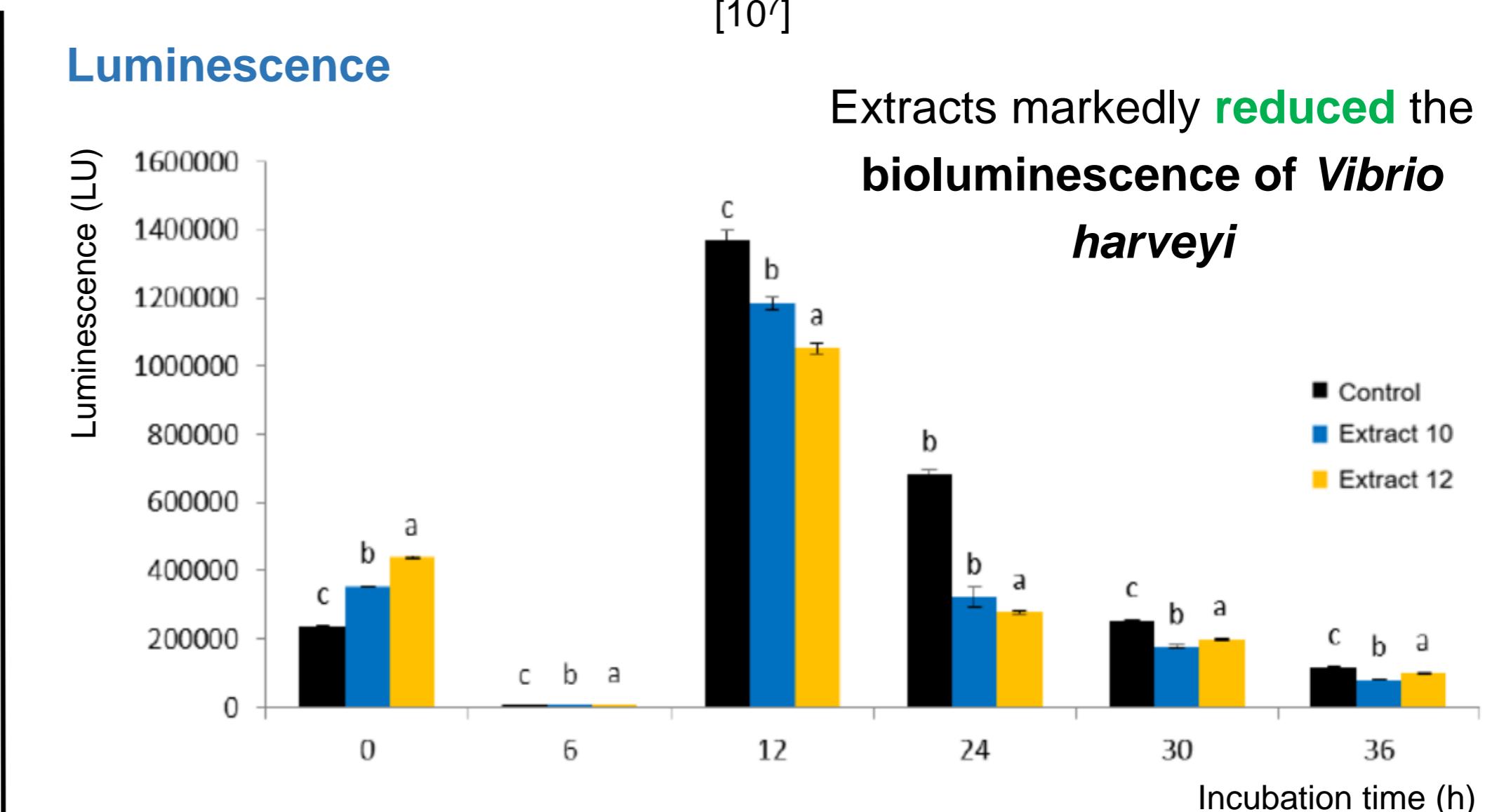
Extracts with better performance



*Vibrio harveyi*  
[10<sup>7</sup>]



Extracts markedly **reduced** the **bioluminescence of *Vibrio harveyi***



Bioluminescence of *Vibrio harveyi* BB120 in MB medium with and without different AEs, Unexposed *V. harveyi* served as control. Values (mean  $\pm$  standard error), at each time point, indicated with different letters are significantly different ( $P < 0.05$ ).

# CONCLUSIES

- In vivo
  - Geen of beperkte toxiciteit voor Artemia
  - Inductie van een “stress respons” in Artemia (phloroglucinol)
  - Interferentie in Artemia - Vibrio model (sommige extracten): verhoogde overleving
  - Reductie van Vibrio bioluminescentie (sommige extracten)

- Acknowledgement:
  - ERANET Marine Biotechnology, FWO: Novel Extraction Processes for muTiple high-value compoUNds from selected Algal source materials (NEPTUNA)

