


From bacteria to fish: ecotoxicological insights into the bioinsecticide Spinosad

Sara Rodrigues^{1*} (sara.rodrigues@fc.up.pt), Sara C. Antunes²

¹ CIMAR/CIIMAR LA - Centro Interdisciplinar de Investigação Marinha e Ambiental, Terminal de Cruzeiros do Porto de Leixões, Avenida General Norton de Matos S/N, 4450-208 Matosinhos, Portugal.

² FCUP - Departamento de Biologia, Faculdade de Ciências da Universidade do Porto, Rua do Campo Alegre S/N, 4169-007, Porto, Portugal.

INTRODUCTION



SPINOSAD (SPI)

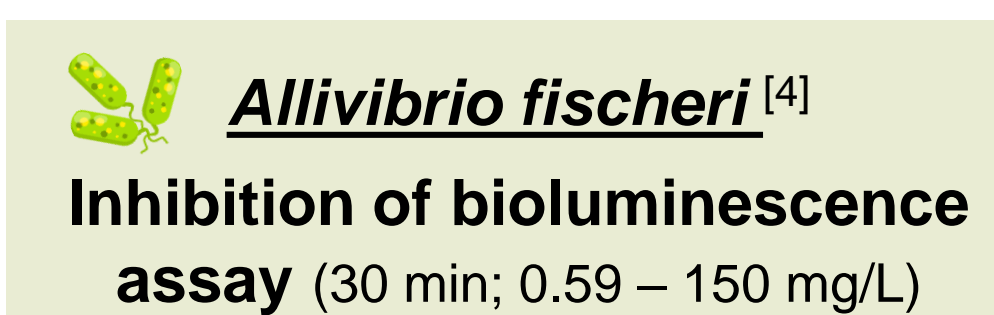
- Natural bioinsecticide derived from the actinobacterium *S. spinosa*^[1] and widely used in agriculture;
- High selectivity toward target pests;
- Low environmental persistence^[1].
- Its increasing application raises concerns about potential toxic effects on non-target aquatic species^[2].

AIM

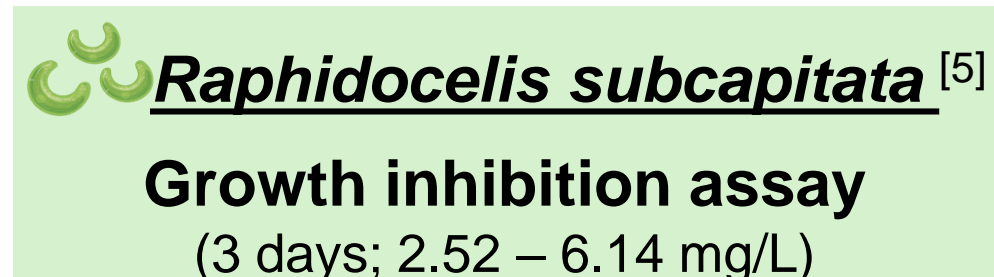
To evaluate the acute toxicity and sub-individual effects of Spinosad (SPI) across multiple aquatic species, representing different trophic levels.

METHODS ^[3]

BIOASSAYS and BIOMARKERS

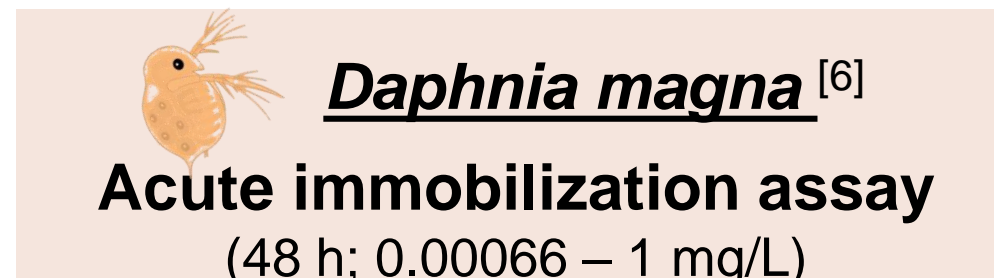


Allivibrio fischeri ^[4]
Inhibition of bioluminescence assay (30 min; 0.59 – 150 mg/L)



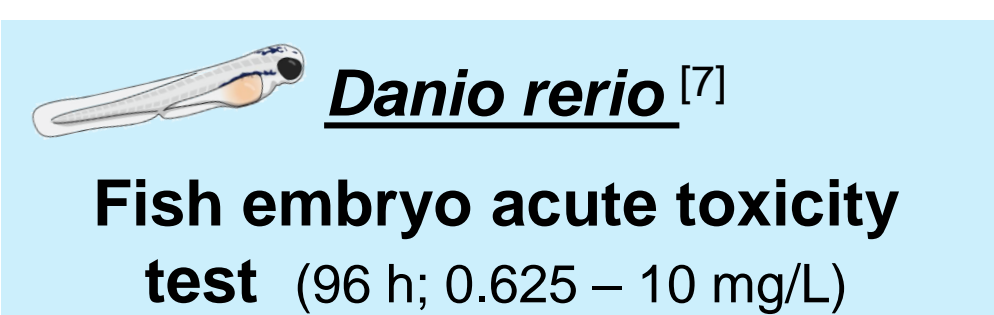
Raphidocelis subcapitata ^[5]
Growth inhibition assay (3 days; 2.52 – 6.14 mg/L)

Sub-individual evaluation
Total Chlorophyll and Carotenoids contents



Daphnia magna ^[6]
Acute immobilization assay (48 h; 0.00066 – 1 mg/L)

Sub-individual evaluation
CAT and GSTs activities, TBARS levels, AChE activity



Danio rerio ^[7]
Fish embryo acute toxicity test (96 h; 0.625 – 10 mg/L)

Growth, Abnormalities, Hatchability

Sub-individual evaluation
CAT and GSTs activities, TBARS levels, AChE activity

ENVIRONMENTAL HAZARD CLASSIFICATION ^[8]

Toxicity score - EU-Directive 93/677/ECC

EC ₅₀ (mg/L)			
Non toxic	Harmful	Toxic	Very toxic
> 100	10 – 100	1 – 10	< 1

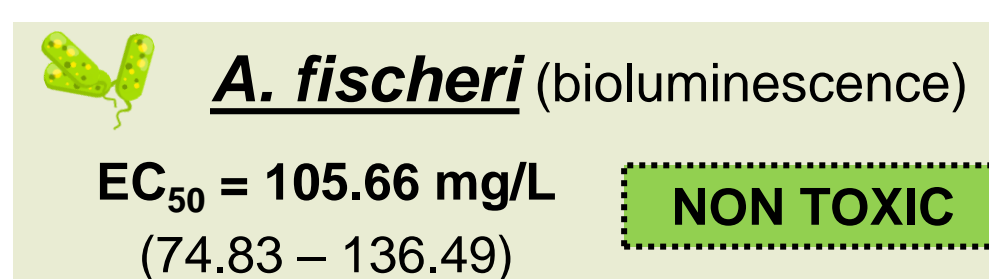
EC₅₀ - Median effective concentration;
NOEC - No Observed Effect Concentration;
LOEC - Lowest Observed Effect Concentration

CONCLUSIONS

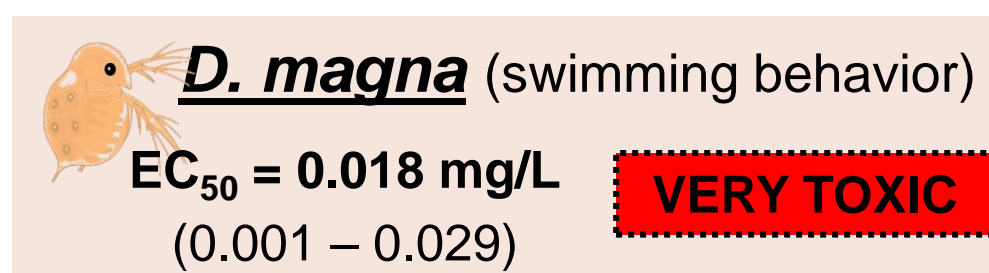
- High toxicity of SPI to aquatic invertebrates namely *D. magna*;
- High potential to disrupt key physiological processes in fish at moderate concentrations;
- More environmental risk assessments of SPI are essential, considering chronic toxicity and sublethal responses, particularly under realistic exposure scenarios to guarantee the health status of aquatic ecosystems.

RESULTS & DISCUSSION

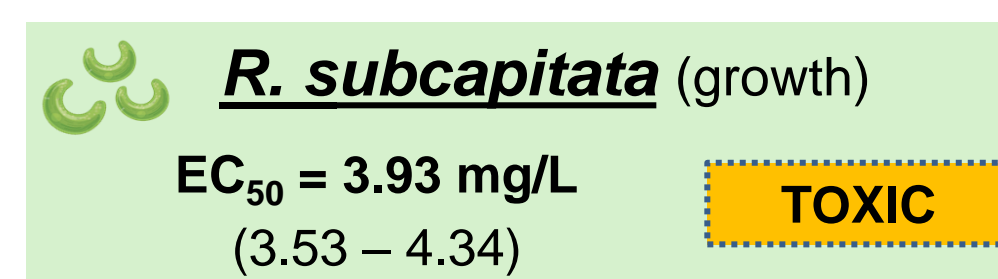
ACUTE TOXICITY



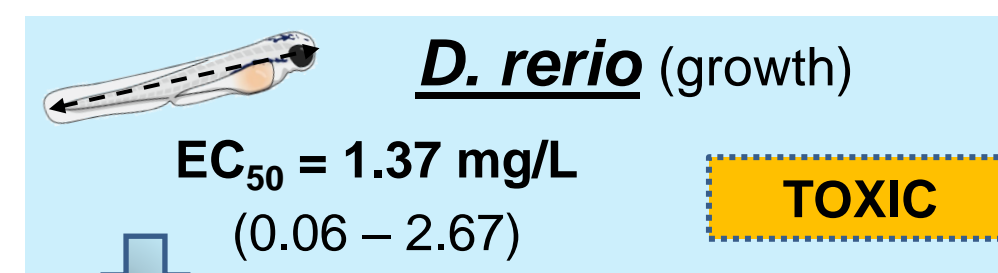
A. fischeri (bioluminescence)
EC₅₀ = 105.66 mg/L (74.83 – 136.49) **NON TOXIC**



D. magna (swimming behavior)
EC₅₀ = 0.018 mg/L (0.001 – 0.029) **VERY TOXIC**



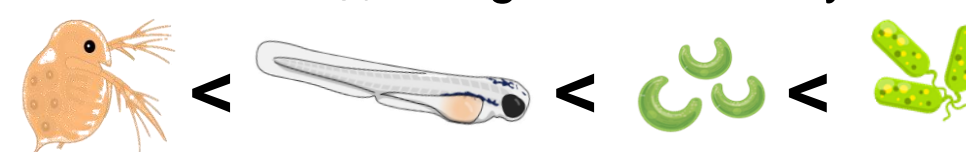
R. subcapitata (growth)
EC₅₀ = 3.93 mg/L (3.53 – 4.34) **TOXIC**



D. rerio (growth)
EC₅₀ = 1.37 mg/L (0.06 – 2.67) **TOXIC**

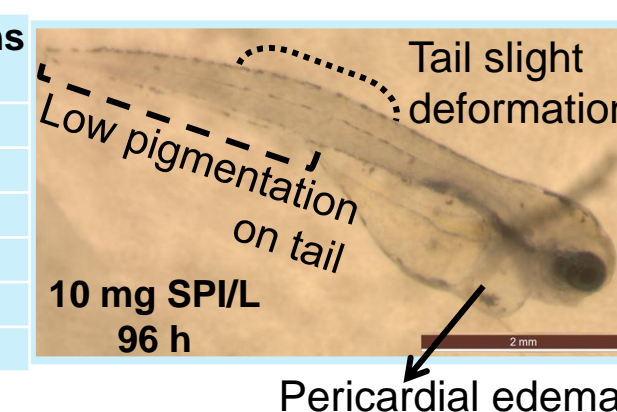
Sensitivity Ranking to SPI

Lower EC₅₀ = Higher Sensitivity



D. magna < *D. rerio* < *R. subcapitata* < *A. fischeri*

SPI (mg/L)	Malformations (%)
0	0
0.625	5
1.25	7.5
2.5	2.5
5	7.5
10	17.5



Tail slight deformation
Low pigmentation on tail
10 mg SPI/L 96 h
Pericardial edema

SPI (mg/L)	Total Chlorophyll content	Carotenoids content
LOEC	4.54	4.54
NOEC	4.11	4.11

SPI caused an impairment of the photosynthetic apparatus, which may be related to oxidative stress or pigment biosynthesis, compromising the photosynthetic efficiency and physiological status of *R. subcapitata*.

SUB-INDIVIDUAL EFFECTS

<i>D. magna</i>				
SPI (mg/L)	CAT activity	GSTs activity	TBARS levels	AChE activity
LOEC	0.0256	1.000	0.0160	0.0102
NOEC	0.0102	0.400	0.0007	0.0041

D. magna exhibited biochemical responses at low concentrations of SPI, indicating high sensitivity:

activation of antioxidant mechanisms and a later phase of the detoxification, pointing to the onset of oxidative stress; lipid peroxidation; early neurotoxic effects.

SPI's toxicity - effects at environmentally relevant concentrations (recommended usage dose for field applications = 500 µg/L)^[2]

<i>D. rerio</i>				
SPI (mg/L)	CAT activity	GSTs activity	TBARS levels	AChE activity
LOEC	10.00	2.50	-	2.50
NOEC	5.00	1.25	10.00	1.25

D. rerio exhibited measurable biochemical responses to SPI, at moderate concentrations:

induction of antioxidant defenses and activation of phase II detoxification pathways, pointing to the onset of oxidative stress; no oxidative damage under the tested conditions; possible neurotoxic effects.

SPI's sub-lethal alterations - detoxification and neural dysfunctions.

