Ecotoxicology TP Course

Concepts, Tests & Biomarkers

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TP2 Photobiology

Activities

PRATICAL COURSE PROGRAM

BEHAVIOURAL

Locomotion capacity
Feeding rates
Attack rates

MORPHOLOGICAL

Growth rates
Cell size
Tissue anomalies
Morphometric and geometry changes

METABOLIC

Photosynthetic activity
Respiratory activity
Sugar/Lipid consumption

MOLECULAR

Enzymatic activity
Membrane peroxidation
Protein oxidation
DNA damage
Gene expression
Metabolite production/consumption

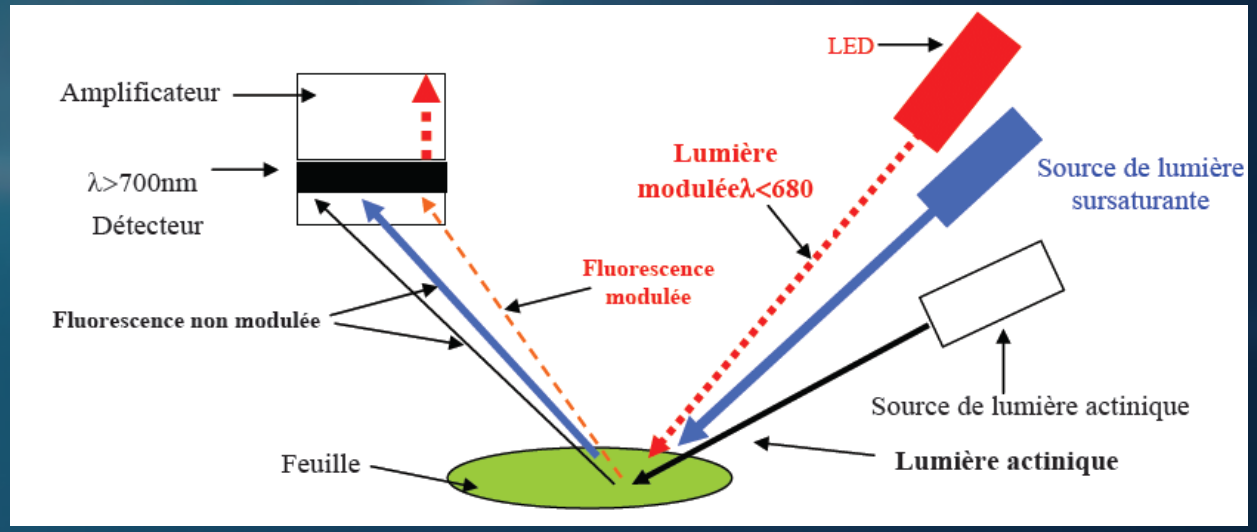
CELL DENSITY AND ECOTOXICOLOGICAL VARIABLES

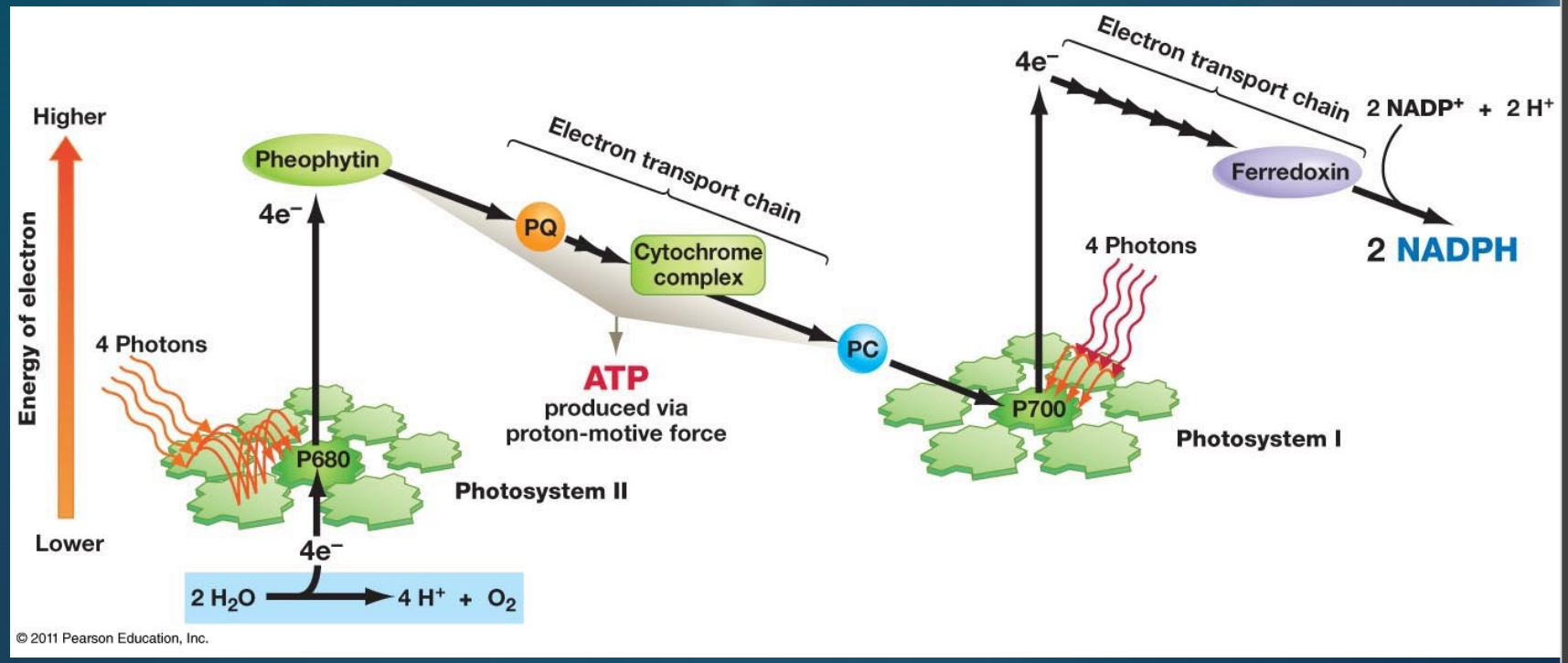
Effects of the test substance in the growth traits and determination of the ecotoxicological doses.

PHOTOCHEMISTRY

Use of remote sensing optical techniques to evaluate the energetic metabolism of the cells under different concentrations of the test dose.


AquaPen
AP 100
MENU
SET


LED
Amplificateur
λ>700nm
Détecteur
Lumière modulée λ<680
Source de lumière sursaturante
Fluorescence modulée
Fluorescence non modulée
Source de lumière actinique
Feuille
Lumière actinique


Higher
Energy of electron
Lower
Pheophytin
4e⁻
PQ
Electron transport chain
Cytochrome complex
ATP
produced via proton-motive force
PC
4e⁻
Electron transport chain
2 NADP⁺ + 2 H⁺
Ferredoxin
2 NADPH
4 Photons
P680
Photosystem II
4 Photons
P700
Photosystem I
4e⁻
2 H₂O → 4 H⁺ + O₂
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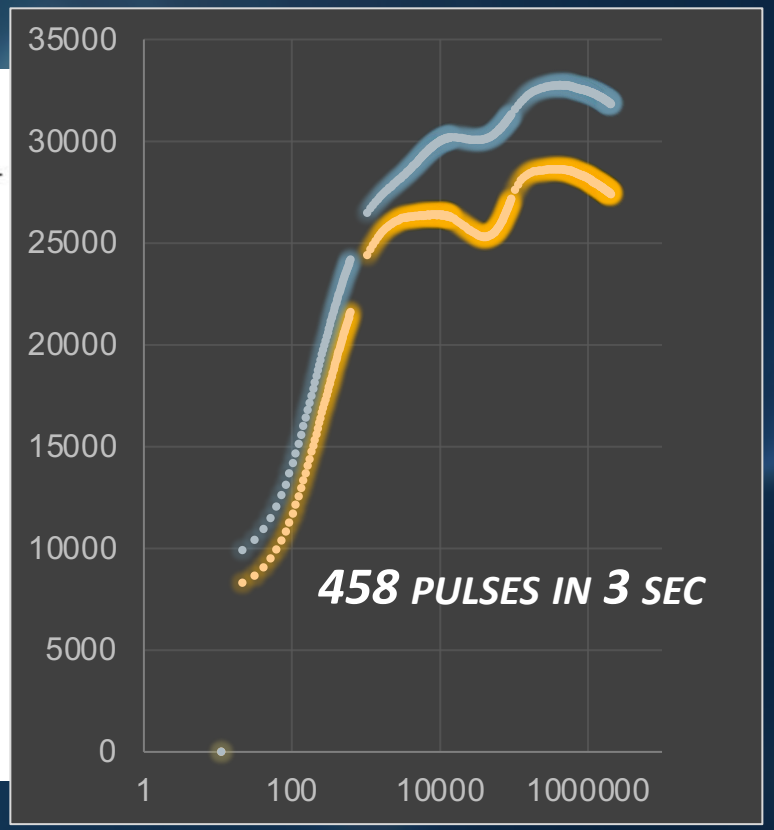
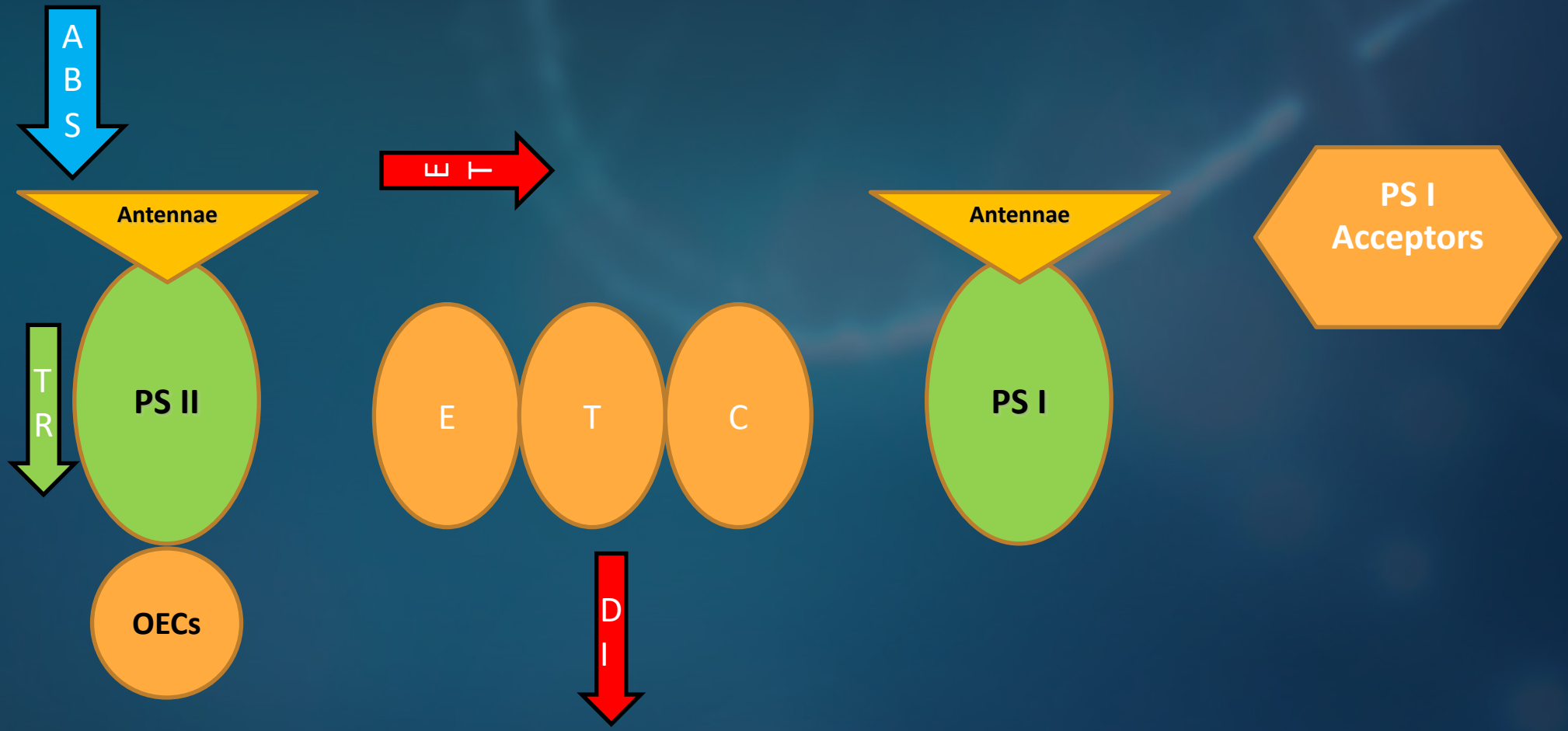

35000
30000
25000
20000
15000
10000
5000
0
1
100
10000
1000000
458 PULSES IN 3 SEC


A
B
S
Antennae
T
R
PS II
OECs
E T
E
T
C
D
I
Antennae
PS I
PS I
Acceptors

Control

10 ug/L Glifosato

250 ug/L Glifosato

500 ug/L Glifosato

PROTOCOL

- Measure basal fluorescence
- Use the OJIP Protocol;
- Calculate the IC50 or LD50 parameters;

Control

10 ug/L Glifosato

250 ug/L Glifosato

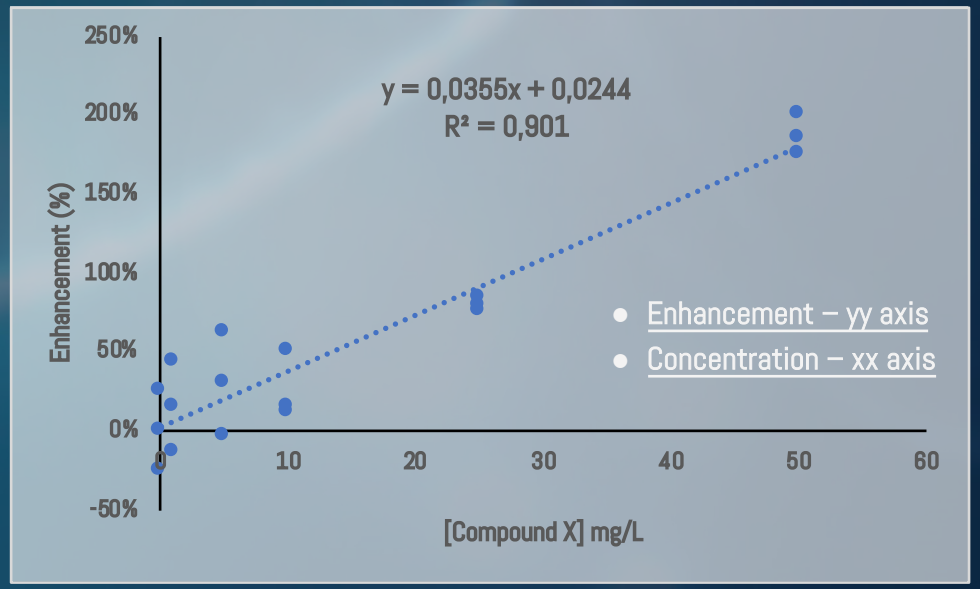
500 ug/L Glifosato

Cell density (cell/mL) = 3429 x Ft – 976200

02 PARAMETERIZATION : Effective Concentration

[Compound X] mg/L	Variable A (a.u.)	Enhancement (%)
0	0,02	0%
0	0,015	-25%
0	0,025	25%
1	0,023	15%
1	0,01725	-14%
1	0,02875	44%
5	0,026	30%
5	0,0195	-3%
5	0,0325	63%
10	0,03	50%
10	0,0225	13%
10	0,023	15%
25	0,035	75%
25	0,037	85%
25	0,036	80%
50	0,06	200%
50	0,057	185%
50	0,055	175%

$$\text{Enhancement (\%)} = \frac{\text{Test} - \overline{\text{Control}}}{\overline{\text{Control}}}$$



Using the linear regression equation calculate the concentration at which the enhancement is 50% (EC_{50})

$50\% = 0.0355x + 0.0244 \Leftrightarrow 0.5 = 0.0355x + 0.0244 \Leftrightarrow 0.5 - 0.0244 = 0.0355x$

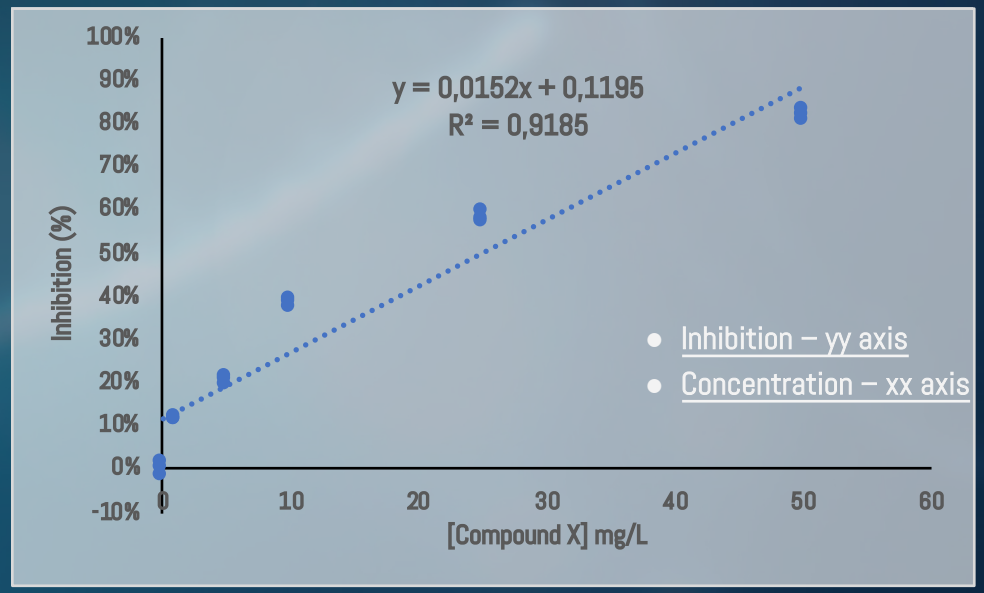
$x = 13.40$ mg/L $= EC_{50}$

Upon the application of 13.40 mg/L the variable A suffers a 50% increase relative to the control.

Concentration

[Compound X] mg/L	Variable A (a.u.)	Inhibition (%)
0	0,02	0%
0	0,015	-25%
0	0,025	25%
1	0,023	15%
1	0,01725	-14%
1	0,02875	44%
5	0,026	30%
5	0,0195	-3%
5	0,0325	63%
10	0,03	50%
10	0,0225	13%
10	0,023	15%
25	0,035	75%
25	0,037	85%
25	0,036	80%
50	0,06	200%
50	0,057	185%
50	0,055	175%

$$\text{Inhibition (\%)} = \frac{\overline{\text{Control}} - \text{Test}}{\overline{\text{Control}}}$$



Using the linear regression equation calculate the concentration at which the inhibition was 50% (IC_{50})

$50\% = 0.0152x + 0.1195 \Leftrightarrow 0.5 = 0.0152x + 0.1195 \Leftrightarrow 0.5 - 0.1195 = 0.0152x$

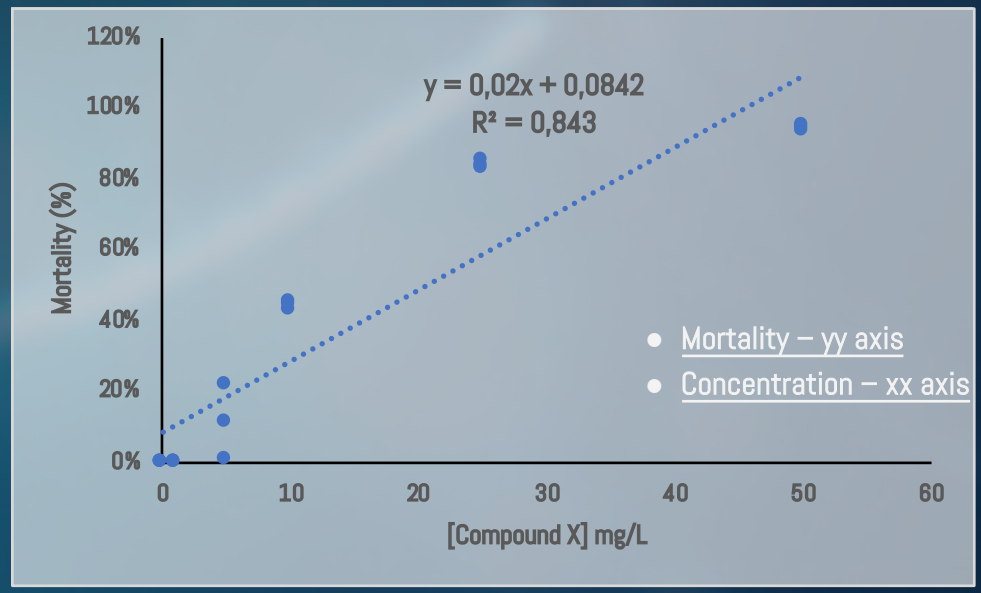
$x = 13.40$ mg/L $= IC_{50}$

Upon the application of 13.40 mg/L the variable A suffers a 50% inhibition relative to the control.

02 PARAMETERIZATION : LETHAL Concentration

[Compound X] mg/L	Variable A (a.u.)	Mortality (%)
0	0,02	0%
0	0,015	-25%
0	0,025	25%
1	0,023	15%
1	0,01725	-14%
1	0,02875	44%
5	0,026	30%
5	0,0195	-3%
5	0,0325	63%
10	0,03	50%
10	0,0225	13%
10	0,023	15%
25	0,035	75%
25	0,037	85%
25	0,036	80%
50	0,06	200%
50	0,057	185%
50	0,055	175%

$$\text{Mortality (\%)} = \frac{\overline{\text{Control}} - \text{Test}}{\overline{\text{Control}}}$$



Using the linear regression equation calculate the concentration at which half the number of initial individuals is dead (LC_{50})

$50\% = 0.02x + 0.0842 \Leftrightarrow 0.5 = 0.02x + 0.0842 \Leftrightarrow 0.5 - 0.0842 = 0.02x$

$x = 15.79$ mg/L $= LC_{50}$

Upon the application of 15.79 mg/L 50% of the individual die.