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IS HERBICIDE TOXICITY ON MARINE MICROALGAE INFLUENCED BY THE NATURAL DISSOLVED ORGANIC MATTER (DOM)?

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Introduction & Objectives

As primary producers, microalgae are the basis of aquatic food webs. Thus, they can be directly affected by herbicides [1]. In their natural environment, these organisms evolve in complex and changing conditions, where various interactions take place. For instance, dissolved organic matter (DOM) may interact with pesticides. Indeed, DOM was shown to interact with pollutants and to affect their transport [2], fate, bioavailability, biodegradation and toxicity on organisms [3].

Material & Methods

This study aimed to investigate whether the natural DOM (from Arcachon bay, Fig.1) influence the toxicity of three herbicides (irgarol (I), diuron (D) and S-metolachlor (S)), singly and in mixtures (M1 and M2), to two marine phytoplankton species: *Chaetoceros calcitrans* (*Cc*) and *Tetraselmis suecica (Ts)*.

Six-day exposure experiments were run using triplicates: each species was exposed to 9 conditions without DOM (Fig. 2) and the same 9 conditions with natural DOM added to the cultures (twice the environmental concentration).

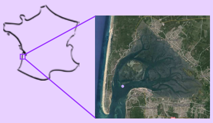

Figure 1 – Arcachon Bay (France)

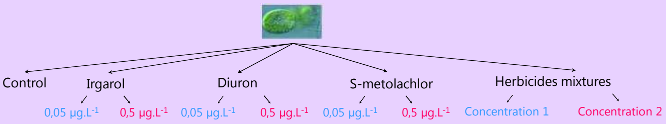

Figure 2 – Treatments applied to each species

The toxic effects were measured on the doubling time (T_D, hours), the photosynthetic efficiency (Y_{eff}), the intracellular ROS relative content using H_2DCFDA and the intracellular lipid relative content using BODIPY (both by flow cytometry).

Results & Discussion

1. Herbicide effects on microalgae

✓ Significant effects were observed for almost all parameters after exposure to I0.5, and M2 for both species (Fig. 3): an increase of the doubling time, corresponding to a strong growth inhibition, a decrease of the photosynthetic yield and a decrease of the relative lipid content. In addition, *Ts* exhibited an increase of relative ROS content.

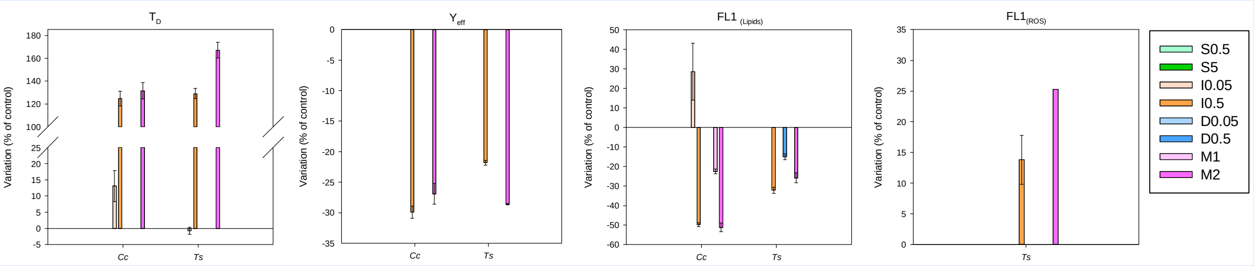

Figure 3 – Significant effects (ANOVA, p<0.05) detected in both species after exposure to S-metolachlor, irgarol and diuron, singly and in mixtures.

✓ Results obtained after exposure of *Cc* to M2 and I0.5 exhibited similar effects: the impacts induced by the mixture 2 might be due to irgarol. In *Ts*, M2 induced a greater growth and photosynthesis inhibition than I0.5 and a greater enhanced H_2DCFDA fluorescence. This higher toxicity would suggest a possible synergistic effect when herbicides are mixed at the highest concentrations. Further experiments are needed to properly investigate this assumption.

2. Influence of DOM on the pesticide effects regarding microalgae physiology

✓ After exposure to herbicides together with DOM, significant effects were observed with I0.05, I0.5, M1 and M2 in *Cc* and with I0.5, D0.05, D0.5, S5, M1 and M2 in *Ts* (Fig. 4).

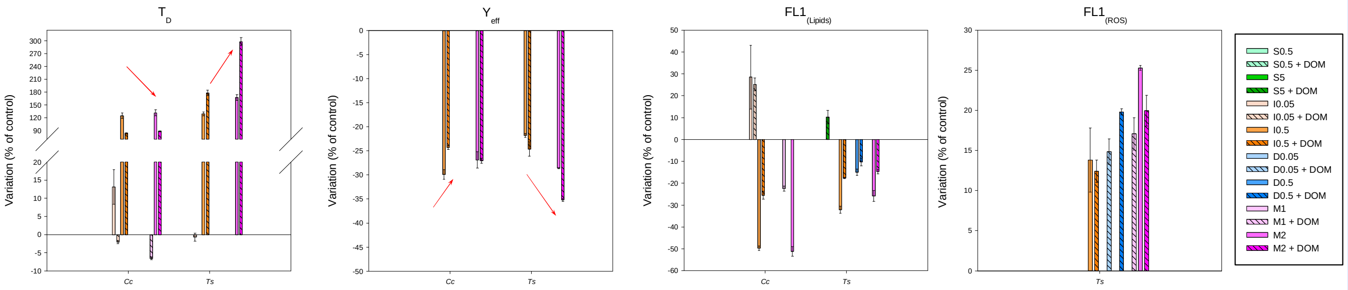

Figure 4 – Significant effects (ANOVA, p<0.05) detected in both species after exposure to S-metolachlor, irgarol and diuron, singly and in mixtures with and without DOM.

✓ The exposure to DOM together with I0.5 or M2 caused quite similar effects on doubling time and photosynthesis efficiency in *Cc*. For *Ts* exposed to I0.5 and M2, results exhibited a much stronger toxicity of M2 compared to I0.5. An effect of D0.05, D0.5, S5 and M1 was also obtained on H_2DCFDA fluorescence, meaning an increase of reactive oxygen species relative content when DOM was added.

✓ When these results are compared to the exposures without DOM, it appears that for *Cc*, the presence of DOM seems to decrease the herbicide toxicity, while it is increased for *Ts*. The decrease or increase of toxicity can be explained by the complexation between DOM and herbicides. Indeed, this complexation can lead to a lower bioavailability of herbicides (because linked to DOM). The difference in herbicide toxicity between the two species in presence of DOM remains to be explained. Analysis of herbicide concentrations, dissolved organic carbon concentration and DOM (in progress) will provide valuable information to improve the understanding of interactions between microalgae, herbicides and DOM in these experiments.

Conclusions

Our results demonstrated the high toxicity of irgarol and herbicide mixtures for two marine phytoplankton species cultivated in absence of DOM. When DOM was added in the culture media, this study showed, in controlled conditions, that natural DOM seems to interact with herbicides and/or microalgae, leading to increased or decreased herbicide effects, depending on the species. Finally, this study demonstrates the importance to consider DOM as a major factor possibly involved in toxicity modulation in the environment.

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