Phaeodactylum tricornutum: new source of eliciting molecules for plant defense and plant health

Coralie Chuberre, Bruno Gügi, Azeddine Driouich, Muriel Bardor, Maïté Vicré

Phaeodactylum tricornutum : new source of eliciting molecules for plant defense and plant health

Coralie Chuberre[1], Bruno Gugi[1], Azzedine Driouich[1], Muriel Bardor[1,2], Maïté Vicré[1]

[1] Université de Rouen Normandie, Structure fédérative de Recherche "Normandie-Végétale", Laboratoire Glyco-MEV EA4358, Centre Universitaire de Recherche et d'Innovation en Biologie (CURIB), 25 rue Lucien Tesnière 76821 Mont-Saint-Aignan Cedex
[2] Institut universitaire de France (I.U.F) 1 rue Descartes, 75231 Paris cedex 05

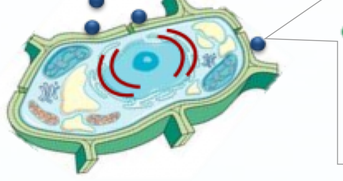
Introduction

Stimulator of plant defense : an alternative to chemical treatments

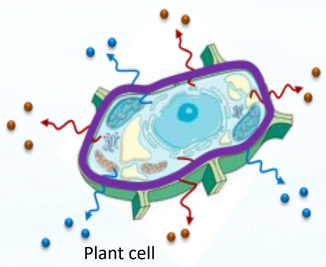
Recently, the use of effective and eco-friendly substitutes to traditional agrochemicals has aroused great interest in agriculture. The development of these new products is based on studies on eliciting molecules, also called elicitors or stimulator of plant defense (SDP), that mimic pathogen aggression to trigger plant defense. Different storage and parietal polysaccharides such as β-glucan, chitin and chitosan oligomers, oligogalacturonides have been reported to induce plant immunity[1]. Among β- glucans, Laminarin extracted from the brown macroalga *Laminaria digitata* is a well-known elicitor of plant defense[2].

Laminarin is extracted from *Laminaria digitata*

1. Laminarin mimic pathogen aggression to trigger plant defense



2. Activation of defense mechanisms

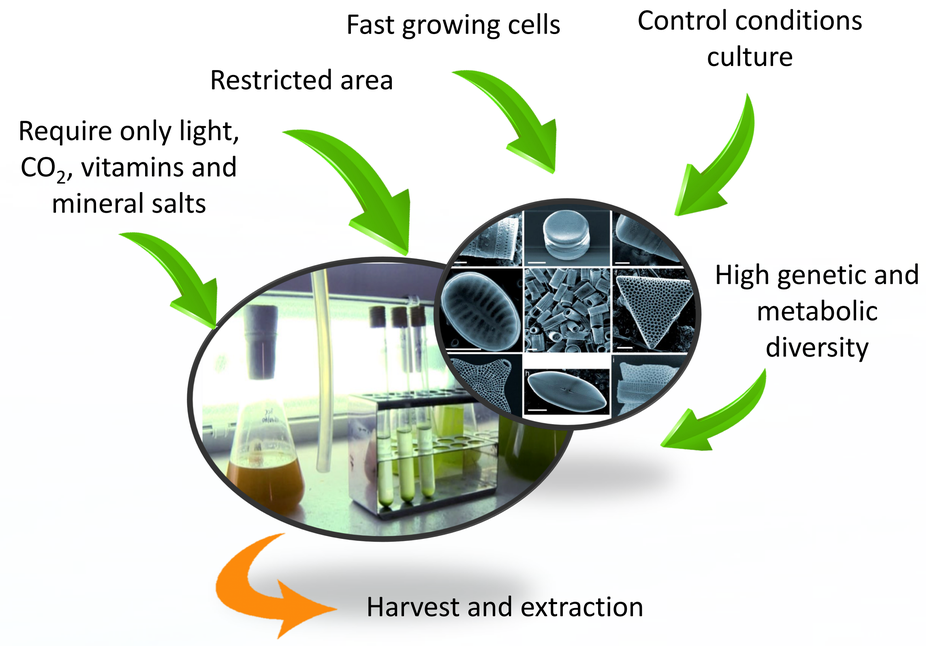


- Cell wall reinforcement
- Antimicrobial compound production
- Enzyme production

3. Plant is protected against future aggression

*Adapted from Goemar Etraining-Vacciplant[4]

Microalgae as a new source of SDP



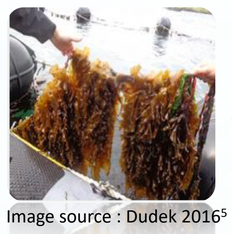
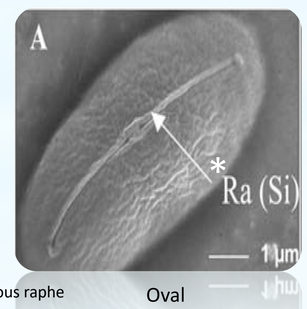


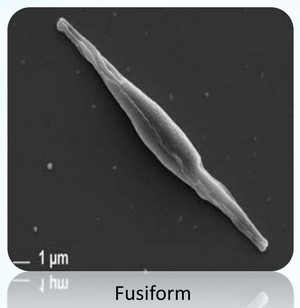
Image source : Dudek 2016[5]

By contrast to macroalgae that require large natural culture areas and take a long time to develop, microalgae grow fast and in controlled restricted areas. They represent a highly diverse group with a wide range of metabolites which represents potential source of eliciting molecules.

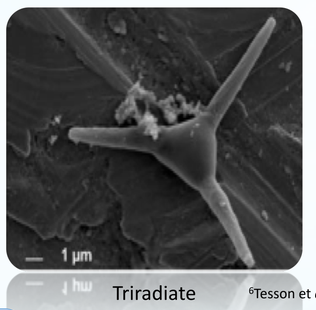
Phaeodactylum tricornutum as a model of microalgae



*Siliceous raphe — Oval



Fusiform



Triradiate — [6]Tesson et *al.* 2009

- Sequenced genome
- **Easy** and **cheap** culture conditions
- **Pleiomorphic cells** depending on environmental conditions[7].
- Production of **allelopathic compounds**[8-9]
- Storage polysaccharide (chrysolaminarin[10]) whose structure is related to laminarin[11]

Promising candidate to identify new elicitor

Arabidopsis root elicitation with *P.tricornutum* crude cell homogenate

P.tricornutum was grown in water complemented with seasalt (33g.L^{-1}), Conway medium (1mL.L^{-1}) and silicate (2mL.L^{-1}). Culture were placed in 16-h-day/8-h-night cycles (50 µE m^{-2} s^{-1}, 19°C).

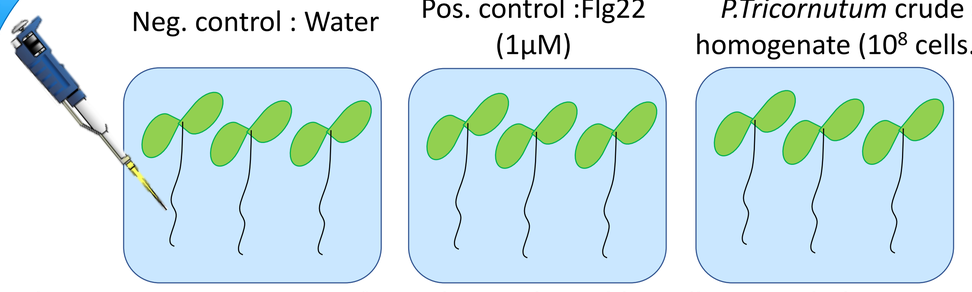
P.Tricornutum cells were harvested after 10 days of growth and ground *via* theFastPrep-24TMsystem (MP Biomedicals) (6x 6,5msx30s)

***P.Tricornutum* crude cell homogenate**

- Neg. control : Water
- Pos. control : Flg22
- Crude cell homogenate

Sterilized *Arabidopsis thaliana* ecotype Columbia seeds are sown onto Murashige and Skoog medium containing 1% (w/v) Bacto Agar (Durand et al., 2009). Plant were grown in 16-h-day/8-h-night cycles (120 µE m^{-2} s^{-1}, 21°C).

Method to evaluate the potential elicitor effect of *P.tricornutum* extract

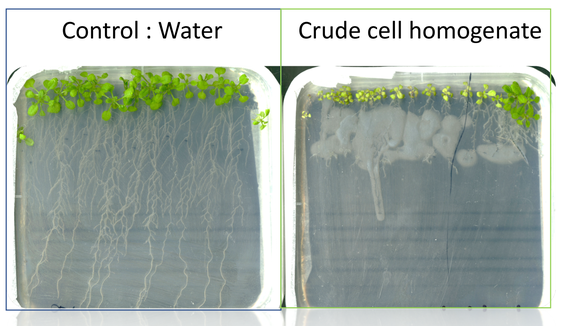


Eliciting treatment was performed on 7 days-old seedlings. Arabidopsis root treated with crude cell homogenate are harvested after different time according to the technique evaluated. Two control were used to evaluated the capacity of crude cell homogenate to induce plant defense. *Flg22 is a famous elicitor of plant defense used as reference in several studies.

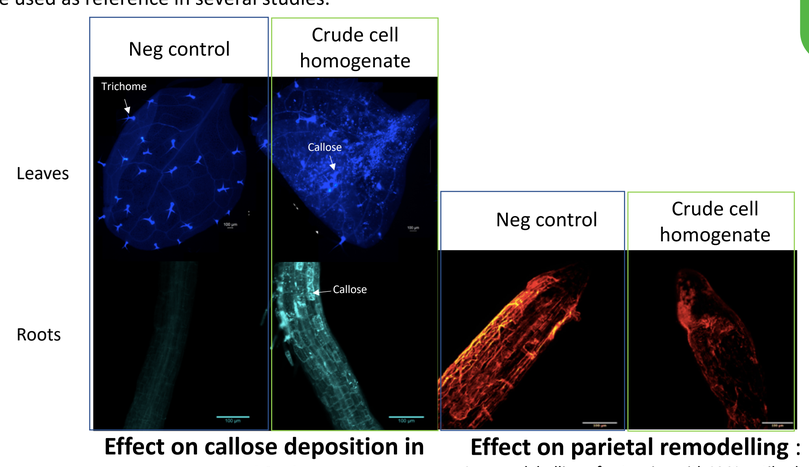
Evaluation of crude cell homogenate efficiency to protect plant

- Effect on plant vigour
- Expression of plant defense genes
- In vitro impact of elicitors on symptom severity and disease progression
- Ability to induce parietal remodelling (Callose and extensins organization)
- *P.Tricornutum* crude cell homogenate
- Effect on bioagressor vigour
- Ability to induce resistance in non treated organ
- Characterization of eliciting molecules

Some results illustrated



Effect on plant vigour :
Root growth was inhibited after microalgae crude cell homogenate for 2 days application on Arabidopsis roots.



Effect on callose deposition in roots and in leaves :
Aniline blue staining showed callose deposition (white arrows) after microalgae crude cell homogenate application for two days.

Effect on parietal remodelling :
Immunolabelling of extensins with LM1 antibody showed a different distribution of labelling when microalgae crude cell homogenate was applied for two days on Arabidopsis roots.

Ability to induce parietal remodelling

Conclusion

Actual results have shown that the crude cell homogenate from *P.tricornutum* induce parietal remodeling and root growth inhibition. However, we need to further investigate the immune response triggered by *Phaeodactylum tricornutum.* To this end, we will study the impact of the elicitor on specific defense markers genes of root defense. Furthermore, a fine structural analysis of the active compound will be performed.

Acknowledgement: The authors would like to thank the Region Normandie (Fellowship for C. Chuberre) and the University of Rouen for their funding.

References:

1. Trouvelot, S. *et al. Frontiers in Plant Science* **5,** (2014).
2. Klarzynski, O. *et al. Plant Physiol.* **124,** 1027–1038 (2000).
3. Norwegian seaweeds (consulted in 2017) : http://seaweeds.uib.no/?art=196
4. Goemar Etraining-Vacciplant (consulted in 2018) : http://etraining.vacciplant.fr/3
5. Dudek 2016, Aberystwyth university, 237p
6. Tesson et *al. (2009)* Botanica Marina. Special issue, Algae Cell Biology. 52, 2, 104-116.
7. De Martino, A. *et al. Protist* **162,** 462–481 (2011).
8. Wang, J. *et al. Scientific Reports* **6,** 37263 (2016).
9. Prestegard, S. *et al.* (2015) *Marine Drugs* **14,** 9
10. Gugi, B. *et al.* (2015) *Marine Drugs* **13,** 5993–6018
11. Caballero, M. A. *et al. Algal Research* **20,** 180–188 (2016).