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Aquafeeds

Optimizing culture of the weissflogii diatom

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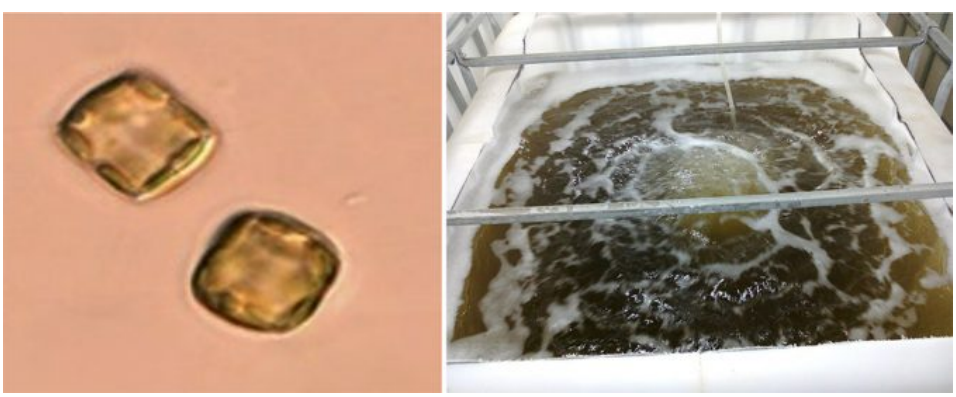
Thalassiosira weissflogii an important input to global shrimp and shellfish larviculture



The diatom *Thalassiosira weissflogii* is important in the aquaculture industry to feed shrimp and shellfish larval stages in hatcheries.

The diatom *Thalassiosira weissflogii* is a widely distributed, unicellular **microalga (https://en.wikipedia.org/wiki/Microphyte)** found in marine, brackish and freshwater environments in many regions of the world. Evidence from molecular genetics suggests that this species of centric diatom is really a species complex and there may be a number of different genetic populations from the eastern and western Atlantic Ocean, California, Indonesia and Hawaii. It is shaped like a short, 4- to 32-μm diameter cylinder, occurring in single cells and in groups embedded in a gelatinous matrix and probably grows better at higher salinities.

The species tolerates poor water quality and it is not toxic, but can sometimes be associated with other microalgal species that can cause red tides and algal blooms. This diatom reproduces both asexually and sexually – the latter can be prompted by external cues like changes in water temperature, light intensity, or day length. It has important uses in the aquaculture industry, where it is used widely to feed shrimp and shellfish larval stages in hatcheries and is considered to be the best microalgae for shrimp larval stages.

In our study, we tested the production of *T. weissflogii* in different photo-bioreactors, to determine the best growing conditions using simple designs and culture conditions.

Testing different photobioreactors

In our study, we tested various photoreactor systems, and here we present our results. *T. weissflogii* is a widely cultured diatom in the global shrimp and shellfish larviculture industries. It is typically cultured at between 1 million and 3 million cells per ml on batch mode and with a growing camera (light path) of around 450 mm. Unlike other species, like *Nannochloropsis gaditana*, which grow very well and at a hundred-fold in 30mm thin layer reactors (vs. conventional 300 mm or 450 mm thick tubular culture reactors), *T. weissflogii* cells bleach to death in very thin-layer (30 mm) photoreactor systems, as their cells divide partially and die, unable to withstand the high photosynthetically active radiation (PAR).

Testing growth of the diatom Thalassiosira weissflogii in containers with walls of different thickness, including 30 mm (left), 210 mm (center) and 450 mm (right).

Technically, it appears that the best growing system for *T. weissflogii* is a cylindrical reactor with 110 m thick walls and a controlled light source capable of adjustments from 60 to 250 μmol photon/m^2/sec, assisted as a time function with respect to growing cell concentrations. With such a set-up, a cell density of 12-15 million cells of *T. weissflogii* per ml can be produced. In principle, an incident photon irradiance (400-700 nm) ranging from 80 to 120 µmol photon/m^2/sec is necessary for the first two days after initial stocking of the diatom, followed by 120 to 160 µmol photon/m^2/sec from day 3 to day 5, and 160 to 200 µmol photon/m^2/sec from day 6 to day 7.

The culture temperature must be kept at 22-23 degrees-C, and air bubbles from aeration provided should be 10 mm in diameter. Before the diatom cells are stocked in the 110 mm-diameter acrylic reactor, the culture must be conventionally grown to a maximum cell density of 3.0 million cells/ml in carboys in a continuous mode for a month and under low nitrogen and low phosphate levels. The photoperiod must be kept at 18 hours light: 6 hours dark, and vitamin B_{12} must be continually dripped in to maintain an available concentration of 0.01 ppm. Providing any more nitrogen will make the *T. weissflogii* cells grow yellowish green.

Also, because *T. weissflogii* tends to stick onto culture walls and to one another, there must be a provision for increasing the air provided (mixing turbulence) for 20 minutes daily in the morning to bring them all into suspension again. Also, fresh seawater must be topped daily to help keep the cells from not clumping together, and this addition should be around 3 percent of the total volume, provided daily using cool, aged (seven days) post-treated seawater.

The best growing system for *T. weissflogii* is a cylindrical reactor with 110-m thick walls.

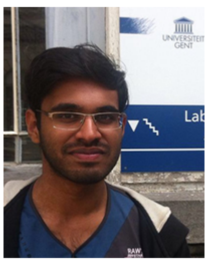
Shading of its cells does not inhibit cell division in *T. weissflogii,* as it has the ability to carry out photosynthesis saturation even at low light intensities. *T. weissflogii* is a nitrate-storing diatom that uses its intracellular nitrate pool for dissimilatory nitrate reduction to ammonium in dark conditions and anoxia, as studied at the Max Planck Institute for Marine Microbiology (Germany) by Dr. Anja Kamp and co-workers.

According to Dr. Paul G Falkowski of Rutgers University (New Jersey, USA), the light saturation index for the growth of *T. weissflogii* has been reported to be approximately 100 µmol photon/m^2/sec. Further, Drs. Costello and Chisholm (MIT, USA) have assessed that 133 µmol photon m^{-2} s^{-1} is the optimal growth irradiance for *Thalassiosira weissflogii.*

Perspectives

Our research shows that the diatom *T. weissflogii* can be successfully cultured semi-continuously and without population crashes for 90 days, and with low nitrogen / phosphate levels (100 ppm N/10 ppm P/3 ppm Urea-N). Cylindrical reactors with 110 m thick walls appear to be optimal for best performance of high density culture of this diatom. The physiological manipulation of cellular light harvesting pigment and cellular photoprotective pigments with high density culture of *T. weissflogii* is still both an art – and science, and thus, more research is needed to fine tune its culture.

Authors



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