Ocean acidification as one of multiple stressors: Response of *Thalassiosira weissflogii* (diatom)

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Nine experiments with *Thalassiosira weissflogii* (Grun.; Fryxell et Hasle 1977; coscinodiscophyceae; CCMP 1053; synonym: *Thalassiosira fluviatilis* Hustedt, 1926) tested the effect of temperature, light, and pCO_2 on elemental composition and growth. Cells were grown at five temperatures (5, 10, 15, 20 and 25ºC) under ambient pCO_2 (Amb.) and low light (LL) conditions (35 µmol s^{-1} m^2). Additionally, cell were grown at 15ºC and 20ºC under future pCO_2 (Fut.) conditions and low light, as well as at 20ºC under higher, but sub-saturating light levels (HL) (65 µmol s^{-1} m^2) at both Ambient and Future pCO_2 conditions.

Cultures were grown semi-continuously in constant-temperature, walk-in incubators, with cell concentrations and pH increasing and nutrients and dissolved inorganic carbon (DIC) decreasing between dilutions. In Ambient treatments the pH fluctuations were targeted to remain between 8.0 and 8.6, whereas in Future treatments the target was between 7.5 and 8.3. All cultures were grown in Fernbach flasks on a 14h-on:10h-off light cycle. Nitrate (NO_3: 58.9 µM), phosphate (PO_4: 3.6 µM) and silicic acid ($Si(OH)_4$: 53.5 µM) prepared as in f/2 were added to seawater based media. The carbonate chemistry was adjusted by adding HCl, $NaHCO_3$, and Na_2CO_3 (closed system approach). Cultures were diluted with fresh media adjusted to the appropriate temperature and pH whenever cell concentrations approached 40,000 to 50,000 cells mL^{-1} or the pH approached 8.6 (Ambient) or 8.2 (Future treatment) to keep cells in exponential growth and the carbonate system within a defined range. Cell concentration (replicate #1 only), dark adapted fluorescence (F_t), quantum efficiency (QY), and pH were determined in all four replicates every 1–3 days, 2–3 hours after the start of the light cycle. On dilution days alkalinity (TA) and frequently dissolved inorganic carbon (DIC) were measured in addition to pH. When the exponential growth rate (µ) was constant (± 0.02 d^{-1}) for at least 8 generations, cell concentrations, the carbonate system, chlorophyll *a* (chl. *a*), dry weight (DW), particulate organic carbon and nitrogen (POC & PON), and transparent exopolymer particles (TEP) were determined in all 4 replicates on four days, either daily or every second day, depending on the growth rate. For methodological details see Passow & Laws, submitted to MEPS.