

Supplementary appendix

Resource limitation of bacterial production distorts the temperature dependence of oceanic carbon cycling

Ángel López-Urrutia, Xosé Anxelu G. Morán

Modelling heterotrophic bacterial metabolism

In order to provide a coherent formulation for the effects of resource availability and temperature on bacterial metabolism we derive a general model for bacterial metabolism. Here, we formulate a set of equations that are used in estimating bacterial growth efficiencies (BGE) and in the prediction of bacterial cell specific respiration (BR_i) and cell specific biomass production (BP_i) from environmental variables like temperature (T , in kelvins) and chlorophyll concentration (chl in mg m^{-3}). According to our conceptual model the total carbon assimilated by a bacteria cell (i.e. the bacterial carbon demand BCD) depends both on temperature and on resource availability following the equation:

$$BCD = b_0 e^{-E/kT} \frac{chl}{chl + K_m} \quad (1)$$

where b_0 is a normalisation constant independent of temperature and resource availability; $e^{-E/kT}$ is Boltzmann's factor where E is the average activation energy for bacterial metabolism and k is Boltzmann's constant; and $chl/(chl + K_m)$ is the Michaelis-Menten functional response of bacterial metabolism to resource availability, where chl serves as a proxy for resource concentration.

Part of this assimilated carbon is devoted to cell maintenance and respired as CO_2 . We have shown that this bacterial respiration is independent of resource concentration but depends on temperature following Boltzmann's factor

$$BR_i = r_0 e^{-E/kT} \quad (2)$$

where r_0 is a normalisation constant independent of temperature. We estimated r_0 and E from our data by regressing the natural logarithm of BR_i against $1/kT$ (Fig 1; Table 1).

The remaining assimilated carbon not used for respiration is used for biomass increase, that is for cell-specific bacterial production

$$BP_i = BCD - BR_i = b_0 e^{-E/kT} \frac{chl}{chl + K_m} - r_0 e^{-E/kT} \quad (3)$$

To estimate the parameters in equation we have to be aware of several limitations of our dataset and of the Leucine incorporation technique used to measure BP_i . First, although the equation above allows for the fact that sometimes net bacterial growth can be zero or negative when available resource concentrations are very low or zero (in which case we would expect a sort period of degrowth before bacteria die), this is not allowed by the Leucine incorporation technique. To measure

BP_i we add a radiolabelled resource (3H-leucine) to the media and hence it is methodologically impossible to measure growth at zero resource concentration. It is therefore not surprising that measured BP_i is always greater than zero. Furthermore, we should keep in mind that chl is just a proxy for resource availability and even if chl was zero a small amount of substrate would still be available for bacterial growth. This lead us to reformulate equation using b_0 and r_0 that differ from the parameters in equations 1 and

$$BP'_i = b'_0 e^{-E/kT} \frac{chl}{chl + K_m} - r'_0 e^{-E/kT} = e^{-E/kT} [b'_0 \frac{chl}{chl + K_m} - r'_0] \quad (4)$$

Second, the correlation between temperature and chlorophyll concentration and the lack of high chlorophyll concentration data at high temperatures cautions against any attempts to fit a multiple non-linear regression to our data. We decided to fit equation in a two step approach. First, we show that the activation energy for BP_i is not significantly different to the activation energy for BR_i (see Fig 2), so we divided BP_i by $e^{-E/kT}$ (where E is the value obtained in the fit of equation ; Table 1) and used non-linear regression following equation

$$BP'_i * e^{E/kT} = b'_0 \frac{chl}{chl + K_m} - r'_0 \quad (5)$$

to estimate parameters b'_0 , r'_0 and K_m . With this estimated parameters it is then straightforward to formulate equation (see Table 1).

The formula that describes the dependence of BGE on resource availability

can be easily derived as

$$BGE = 1 - \frac{BR_i}{BP'_i + BR_i} = 1 - \frac{r_0 e^{-E/kT}}{e^{-E/kT} [b'_0 \frac{chl}{chl+K_m} - r'_0] + r_0 e^{-E/kT}} \quad (6)$$

and symplifying we arrive to

$$BGE = 1 - \frac{1}{\frac{b'_0}{r_0} \frac{chl}{chl+K_m} - \frac{r'_0}{r_0} + 1} \quad (7)$$

if we substitute the parameters in equation 7 with those fitted using equations and 5 we arrive to the equation presented in Table 1 and represented in Fig 3C.