Supplementary information

Analysis of additional pigments

Two of the experiments were carried out with *Rhodomonas salina*, a Cryptophyte that, additionally to Chl-*a*, contains phycobiliproteins as accessory pigments. Phycoerythrin emits fluorescence in the orange portion of the spectrum when is excited with blue light. Hence, for each treatment of the experiments 7 and 8 (Table I in the main text), measurements of single cell orange fluorescence were acquired with FlowCAM. Excitation illumination consisted of a blue laser fan of 488 ± 0.04 nm, and fluorescence was measured as the emitted light passing a 575 nm band-pass filter (30 nm) and reaching a photomultiplier tube (PMT 2). The maximum value reached by the voltage pulse generated when the fluorescent light reaches the PMT (pulse peak) was considered as a proxy for the phycoerythrin content of the cells. Hence, although a conversion between the intensity of the fluorescence and the pigment content was not performed, the orange fluorescence was used to explore the changes in the content of accessory pigments during the incubations.

The measurement of fluorescence on a single-cell basis allowed us to explore the size dependence of pigment content within a single population. To analyse the size dependence of accessory pigments content as a function of irradiance within each single population, the fluorescence of each single cell was plotted against cellular biovolume in a log-log scale. As an example, the five irradiance treatments in experiment 7 are shown in Figure SI (increasing irradiance from panels A to E). The distributions of biovolume and fluorescence per cell were log-normal, as indicated by the bar plots in the x and y axis. Since the data clouds presented high dispersion and several outliers, we applied robust linear regression to obtain the size scaling exponent. Although the slope of the regression models were significantly different

from zero in most of the treatments (p-value<0.05), body size explained a small proportion of the variability in pigment content (between 3 and 12%).

Considering a single species, the size scaling exponent tended to decrease in light saturated conditions. In Figure SII we present the variation of the size-scaling exponent as a function of irradiance for the two experiments with *R. salina*. Both experiments showed the pattern of decrease of the size-scaling exponent at increased growth irradiance levels, although only in the experiment 7 the tendency was significant (p-value<0.05, n=4). This pattern was the expected from photo-protective pigments. The concentration of these pigments increased in light saturated conditions, but small cells could increase their concentration more than larger cells that were constrained by smaller surface to volume ratio. This turned in a flatter size scaling exponent, an opposite trend to that found for the light harvesting pigments, such as Chl-*a*. Although it is not possible to generalize on the basis of such scarce information, it appeared that the different patterns of light-harvesting and photo-protective pigments can be depicted thanks to the measurement of their different fluorescent signatures on a single-cell basis.

Figures

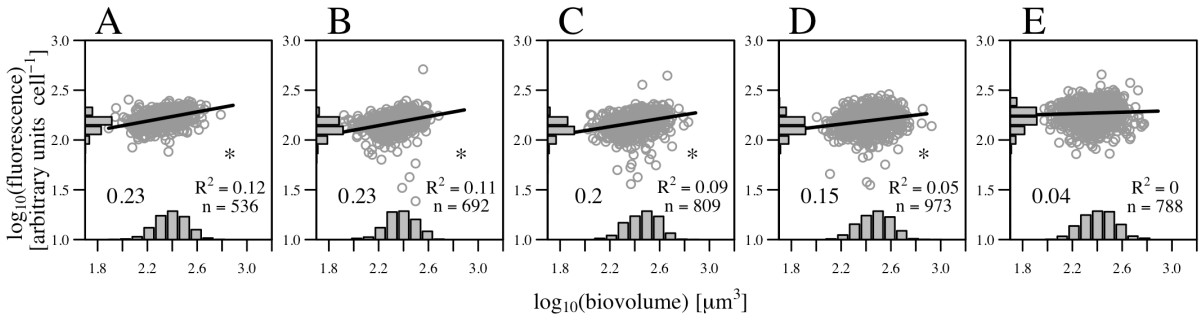

Figure SI. Intra-specific size scaling of orange fluorescence per cell. Population size scaling in the five increasing (**A** to **E**) irradiance treatments of experiment 7 (*R. salina*).

The size scaling exponent was obtained through robust linear regression; the slope, R^2 of the relationship and the number of cells analysed are shown in each panel. The asterisk in each panel indicates that the relationship was significant (p-value<0.05). Bar plots attached to the x and y axis show the distribution of cell biovolume and orange fluorescence respectively.

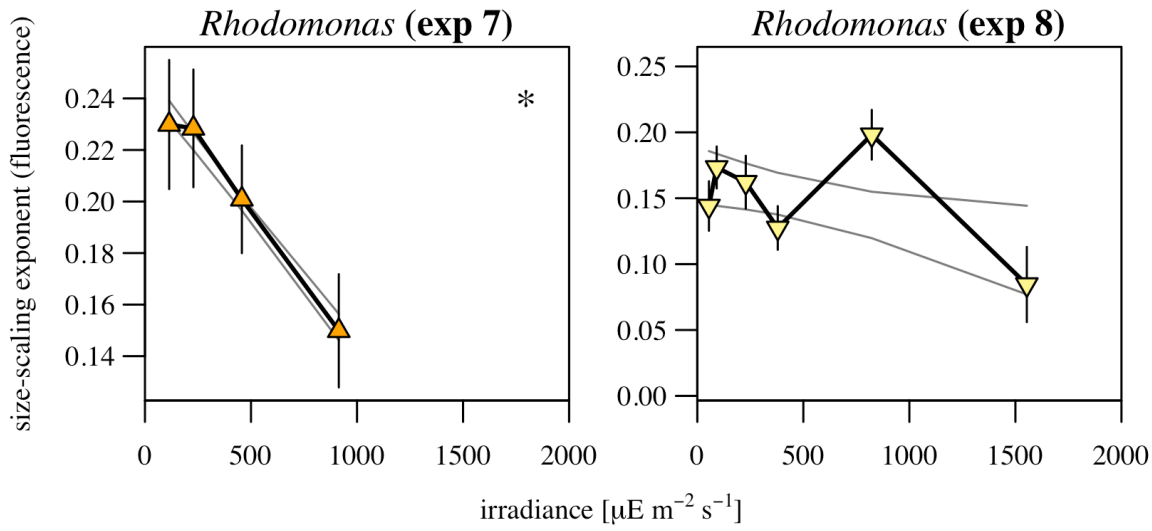

Figure SII. Change of the intra-specific size scaling with light. Intra-specific size scaling exponent for orange fluorescence (arbitrary units) per cell, assimilated to phycobiliproteins-like pigments, as a function of growth irradiance for the two experiments carried out with *R. salina*. Size scaling exponents were calculated from robust linear regression with standard deviation show in vertical bars. The asterisk in each panel indicates that the relationship was significant (p-value<0.05).