

Noga Stambler · Zvy Dubinsky

Corals as light collectors: an integrating sphere approach

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Abstract An integrating sphere was used to estimate the fraction of the incident quantum flux absorbed by a coral colony placed within it. This method allows one to examine the *in vivo* light absorption of intact coral colonies. We used this method to study effects of colony morphology, size, and photoacclimation status on the light harvesting efficiency by the zooxanthellae. Light absorption per unit of coral surface area decreased with increase in colony size, with a clear effect of different coral morphologies. In branched colonies, shading among branches reduced the absorbed light per unit area and per zooxanthellae. Photoacclimation to low light resulted in increased cellular chlorophyll concentrations in the zooxanthellae. In shade acclimated colonies, areal chlorophyll concentrations increased significantly, leading to more overlap among the optical cross-sections of pigments within cells and mutual shading among cells. These “package effects” showed up as a decrease in the *in vivo*, chlorophyll-*a* specific, spectral average, effective optical cross-section, a^* . An integrating sphere is a useful tool for collecting optical information on corals.

Keywords Corals · Light · Absorbed · Integrating sphere

Introduction

Coral reefs are roughly limited to the euphotic zone in which the light is less than 1% of its subsurface value (Wells 1957; Stoddart 1969; Dustan 1979, 1982; Fricke and Meischner 1985; Wyman et al. 1987). In the oligo-

trophic “blue deserts” of tropical seas, where coral reefs dominate coastal zones, the euphotic zone extends to depths of up to 120 m. The decrease in light shapes the depth distribution of reefs. The number of hermatypic coral species (Achituv and Dubinsky 1990) and the calcification rate of reef-building corals (Barnes and Chalker 1990) follow courses parallel to that of light attenuation. Beyond the effects of region, time, depth, and water transparency the intensity and the spectrum of the light that reaches any coral also depends on its specific location and orientation. Corals growing in a cave or in shade receive light as compared to exposed ones.

The quantity and spectral distribution of light to which corals are exposed depends on reef latitude, season, and time of day. The light field depends on the vertical attenuation of light with depth. The effect of depth on light, or formally the vertical attenuation coefficient (Kirk 1994), depends on the light absorbing and scattering properties of water itself. It also depends on all particles and dissolved substances in it, the main ones being phytoplankton cell pigments, suspended sediment, and dissolved and colloidal organic compounds including “yellow substance”. The exponential decrease in light intensity with depth is accompanied by changes in the spectral distribution of the light field since all of the above listed substances have characteristic wavelength-dependent optical properties

Corals absorb a variable fraction of the total radiation flux incident on them, depending on their shape and the wavelength-specific absorptance: the remaining light will be scattered or reflected (Gates 1980).

Most hermatypic corals harbour in their tissues symbiotic algae, the zooxanthellae. These symbionts belong to the Dinoflagellata (Dinophyceae), and, as such, contain distinctive photosynthetic pigments, mainly chlorophyll *a*, chlorophylls *c*₁, *c*₂, and carotenoids such as peridinin. The composition of photosynthetic pigments is reflected in the corals specific spectral absorption, which is also related to the action spectrum of photosynthesis (Halldal 1968; Scott and Jitts 1977).

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N. Stambler (✉) · Z. Dubinsky
Faculty of Life Sciences, Bar-Ilan University,
Ramat Gan, 52900, Israel
E-mail: stambln@mail.biu.ac.il
Tel.: +972-3-531-8283
Fax: +972-3-535-1217

Additionally, coloured water-soluble pigments in the animal tissue, as well as the mycosporin-like amino acid (MAA) S-320 and related compounds, absorb UV radiation (Shibata 1969; Dunlap and Chalker 1986). These compounds protect both the host and the symbiotic algae from UV radiation damage. The amount of the UV absorbing pigments in a variety of zooxanthellate hosts increases towards the surface (Shick et al. 1995; Banaszak et al. 1998; Lesser 2000; Shick and Dunlap 2002)

Shibata and Haxo (1969) measured the absorption spectra of zooxanthellae and endolithic algae found in the skeleton of the coral *Favia pallida* Dana. Halldal (1968) cited unpublished data from Shibata and Haxo showing that in the coral *Favia*, an average of only 0.1% of the light in each wave-length penetrates the layer of the zooxanthellae, animal tissue, and skeleton to reach the endolithic algae. Halldal (1968) and Shashar and Stambler (1992) found that this amount of light may be sufficient for active photosynthesis and to maintain the endolithic algae.

To fully characterize the photosynthetic process of the zooxanthellae, one has to know the amount and spectral distribution of the PAR (photosynthetically available radiation) (Morel 1978) absorbed by the coral colony, in addition to the carbon assimilated, or oxygen evolved over the same time interval. These data may be used to calculate the quantum yield of photosynthesis (Dubinsky 1980; Dubinsky et al. 1984, 1990; Wyman et al. 1987). The efficiency of photosynthesis is of interest, since it is related to both the photoacclimation and nutrient status of the coral-zooxanthellae association, as well as being a potential indicator of stress. In the present study we attempted to measure coral absorption.

There are some indirect ways to calculate the amount of the absorbed light of zooxanthellae, all of which are based on absorption measurements of isolated zooxanthellae. Dustan (1982), in his pioneering study found that the whole-cell absorption of zooxanthellae of the coral *Montastrea annularis* increases with depth. His measurements were done on frozen, isolated zooxanthellae that were suspended in a small volume of seawater with 0.1 g CaCO₃. Working with zooxanthellae collected on filter paper Dubinsky et al. (1984), Wyman et al. (1987), and Lesser et al. (2000) were able to compare the absorption of PAR by zooxanthellae from different light environments. In these studies, it was found that in the course of photoacclimation to low light, the concentration of cellular chlorophyll and other light-harvesting pigments increases up to ten-fold compared to that of high-light acclimated cells. However, at the same time the chlorophyll-specific absorption coefficient, a^* , decreases due to inter- and intracellular “package effects” (Dubinsky et al. 1984, 1986, 1990; Sukenik et al. 1987, Berner et al. 1989). That absorption coefficient is defined as the in vivo absorption coefficient of algal cells, normalized to their chlorophyll *a* content. The value of a^* may be determined for each wavelength,

or be averaged over the whole visible spectrum (the PAR range). For definitions see Dubinsky (1992) or Schanz et al. (1997). Packaging effect results from the photoacclimative changes in cellular pigmentation, as well as from nutrient induced changes in zooxanthellae densities (Dubinsky et al. 1984, 1990).

Obviously, there are some disadvantages inherent in the use of isolated zooxanthellae. The absorption of the zooxanthellae in vivo depends on the micro-scale environment within the host, such as whether they are in the tentacles or in parts deep within the calyx, the thickness and pigmentation of the surrounding host tissue, and whether their location is at the proximal or distal end of coral branches. For instance, Schlichter et al. (1986) reported that in the coral *Leptoseris fragilis* there is a difference between the absorption spectra of isolated zooxanthellae and that of zooxanthellae within the host tissues. These authors suggest that deep-sea corals provide the zooxanthellae with additional light by transforming light of short wavelengths into wavelengths suitable for photosynthesis, and by directing light towards algal cells by specialized skeletal structures. In addition, measurements of absorption by algae collected on filters are likely to be overestimated due to “path amplification” effects, as shown by Kiefer and Soohoo (1982) and Bricaud and Stramski (1990) in their studies of free-living phytoplankton.

Integrating (or Ulbricht) spheres have been used to measure the fraction of incident quantum flux that is absorbed by plant material placed into the sphere (Rabideau et al. 1946; Welschweyer and Lorenzen 1981; Haardt and Maske 1987) and even to estimate whole-plant chlorophyll content (Macdowall 1983).

Our aim was to use the integrating sphere methodology to study whole coral colonies in order to examine the following:

1. What is the effect of colony size on light absorption?
2. Are there differences in light-absorbing properties between massive and branched corals that affect their performance under different ambient light intensities?
3. What is the efficiency of photoacclimation of the zooxanthellae in optimizing light harvesting?

Materials and methods

Collection

Colonies of five species of corals: *Acropora* sp ($n=9$), *Stylophora pistillata* ($n=20$), *Pocillopora damicornis* ($n=7$), *Favia* sp. ($n=7$), *Fungia* sp. ($n=8$), and of the hydrocoral *Millepora dichotoma* ($n=8$), were collected in Eilat (Red-Sea), from depths of 1–3 m. They were placed in aquaria with running seawater. All measurements were carried out a few hours after collection at the Interuniversity Institute of Eilat (The H. Steinitz Marine Biology Laboratory), Israel. Nine high-light and ten low-light colonies were collected from shallow

water locations, in order to minimize the effect of various depth-related environmental factors other than light. The low-light adapted colonies were taken from shaded crevices and under ledges (1–10% of surface light). Whenever large colonies were subdivided into fragments, for comparison among species, they were collected from the same light level, and were of similar size.

Integrating sphere

The integrating sphere was built of two vertically hinged halves, 18 cm in diameter, painted white with Barium Sulphate (BaSO_4) based reflectance paint. The coral colonies were placed in a beaker with seawater on a pedestal in the centre of the sphere. Light was introduced into the sphere by a fibre-optic low-noise illuminator model 9741-52, (Cole-Parmer Instrument Co., Chicago), equipped with a 150 W quartz-halogen bulb, and diffused by a baffle. That arrangement, ascertaining an isotropic light field, minimizes effects due to the glass beaker and that water was only within the beaker and not in the entire sphere. Furthermore, by subtracting tissue-stripped values from those of the live corals, potential bias would have been minimized.

Light from the integrating sphere was analysed with a Techtum Quantaspectrometer QSM-2500 (TQ), Uppsala, equipped with an optic fibre collector, which was inserted into the sphere behind a second baffle. The two baffles provided an evenly scattered light field within the sphere.

The spectrometer was interfaced with a computer for both scan control and data acquisition and processing (Dubinsky et al. 1990). The complete integrating-sphere set-up is outlined in Fig. 1. Two separate programs written in Turbo Pascal called SCAN and SPEC, respectively, controlled the data acquisition and analysis. The scan program records a spectrum from a single real-time sweep between 400–750 nm, at 1.5 nm intervals. The SPEC program provides a complete analysis of the set of spectra recorded by SCAN (Dubinsky et al. 1990).

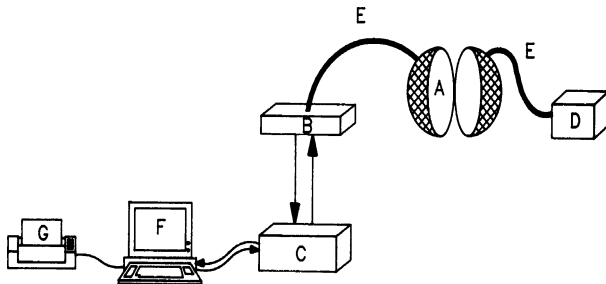


Fig. 1 Integrating sphere set-up. *a* Integrating sphere. *b* Quanta spectrometer. *c* Control box. *d* Illuminator. *e* Fibre optic. *f* Computer. *g* Printer

Measurements of absorption

For non-branched corals whole colonies of *Favia* sp. and *Fungia* sp. were used. Colonies of the branched species *P. damicornis*, *S. pistillata*, *Acropora* sp., and of the hydrocoral *M. dichotoma* were subdivided before use. In all cases the surface area of the colonies, or branches ranged from 5 to 200 cm^2 . Each sample was scanned two to five times, and the values presented are means of all scans. Since these replicate scans gave virtually identical results no deviations are given. After measurements, the tissue was removed by a seawater jet from a WaterPik (Johannes and Wiebe 1970). The bleached coral skeleton was placed in the integrating sphere, while maintaining the previous orientation, and re-scanned. For the study of the relation of absorbed light to colony surface area, additional branches of various sizes from the same, subdivided colony were used to obtain the desired area range.

Q_{abs} , or the amount of light energy absorbed by the living tissue, is the difference between the spectral irradiance measured in the integrating sphere, (Fig. 2) with the coral skeleton, I_s , and that measured with the intact coral, I_c (Eq. 1).

$$Q_{\text{abs}} = I_s - I_c \quad (1)$$

The percent of light absorbed was calculated (Eq. 2)

$$\% \text{abs} = 100 * \frac{I_s - I_c}{I_s} \quad (2)$$

The spectrum of the light absorbed by the corals, as measured in the integrating sphere, reveals features typical for photosynthetic pigments such as a chlorophyll *a* absorption with a peak at 675 nm (Fig. 2).

For every discrete wavelength, I_c/I_s depends on the spectral absorbance of the living tissue (Gates 1980). The absolute number of quanta absorbed by the living coral, and their spectral distribute, depends on the spectral distribution of the scalar irradiance to which it

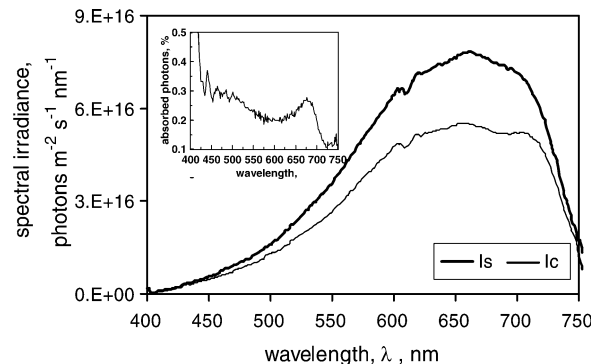


Fig. 2 The spectral irradiance measured in the integrating sphere, for intact coral (I_c) and for the coral skeleton (I_s). The difference between these two curves represents the light absorbed by the living tissue. The spectrum of the light absorbed by the tissue is shown in the small insert

is exposed, and on the spectral absorptivity of the tissue (Gates 1980; Dubinsky et al. 1986).

Biomass parameters

In order to correlate the absorption and biomass data, the following parameters of each coral colony were measured: colony surface area, volume of homogenate, and the concentrations of algae and chlorophyll *a* per unit volume of homogenate. From these, areal concentrations of zooxanthellae, of chlorophyll *a*, and cellular chlorophyll *a* content in zooxanthellae were calculated.

Surface area of branched colonies was measured on a Delta-T device area meter (Decagon Devices Ltd.), after the waterpiked skeleton was broken up, to avoid overlap between branches. The projected area was multiplied by π , assuming a sub-cylindrical geometry (Falkowski and Dubinsky 1981). The surface area of hemispherical and flat corals was measured by the aluminium foil method (Marsh 1970). The area of the aluminium paper was measured on Delta-T device area meter. Tissue homogenate was prepared by quantitatively removing all tissue with a WaterPik (Johannes and Wiebe 1970). Density of zooxanthellae in the homogenate was determined by microscopic examination using a haemocytometer, and chlorophyll *a* concentration was measured spectrophotometrically on a Beckman DU-6 spectrophotometer, after extraction with 90% acetone, and calculated according to the trichromatic equations of Jeffrey and Humphrey (1975).

The absorbed light per square centimetre, zooxanthellae, and chlorophyll *a*, were calculated and compared for the different corals. Unlike in a solution, the absorption of native chlorophyll *a* depends on the fine architecture of the cellular light-harvesting apparatus and varies with taxonomic affiliation, cell geometry, photoacclimation, and nutrient status. This property is defined as a^* , the in vivo optical cross-section, and when normalized to chlorophyll *a*, as the in vivo optical cross-section “chlorophyll-*a* specific”. This parameter is wavelength dependent and may be averaged over the visible spectrum (Morel and Bricaud 1981; Kiefer and Mitchell 1983; Dubinsky et al. 1984, 1990; Dubinsky 1992; Schanz et al. 1997). This averaging allows one to estimate the total amount of light available for photosynthesis. The spectral-averaged values of a^* of the zooxanthellae were calculated from the integrating sphere results. For comparison, we also determined a^* using the filter paper method following the procedure of Dubinsky et al. (1984). We used freshly isolated zooxanthellae, separated from the homogenate by centrifugation and resuspension in filtered seawater. Different amounts of the algae were collected by filtration on Whatman GF/C filters. The absorbance and chlorophyll *a* concentration of these filters were determined. From an exponential regression of absorbance against areal chlorophyll *a*, the parameter a^* is calculated as described in Dubinsky et al. (1984).

Results

As might be expected, the amount of absorbed light measured in the sphere was correlated with colony size, since the larger the colony, the more possible light paths were blocked. When small coral pieces were placed in the integrating sphere, more “remaining”, unabsorbed, light was measured, than with large ones (Fig. 3, $r^2 = 0.99$). In this figure, since the illuminating beam intensity was not changed, the sum of the non-absorbed light and the absorbed fraction remained constant. When normalized to surface area, the amount of light absorbed per unit area decreased with the increasing coral size. Such a decrease was found in the branching coral *Acropora* sp. ($r^2 = 0.84$) and the flat hydrocoral *M. dichotoma* ($r^2 = 0.97$) (Fig. 4a, b). In both cases we used one colony that was broken into several fragments of different sizes. All of these fragments were measured, and the data are included in Figs. 4 and 5. For both species, a higher percent of the total incident light was absorbed by larger colonies; whereas, the opposite was true for the areal percent absorption which decreased with colony size (Fig. 5a, b). However, the inverse relation between coral surface area and light absorbed is polynomial for *Acropora*, $r^2 = 0.84$ (Figs. 3 and 4a); whereas, it is linear in *Millepora* $r^2 = 0.97$ (Fig. 4b).

Percent absorbed photons for colonies of several species, the branched *Acropora* sp., *S. pistillata*, and *P. damicornis*, the flat *M. dichotoma* and *Fungia* sp., and *Favia* sp. as a spherical coral, can be seen in Fig. 6. That figure is based on over 10 colonies of each species. Except in the case of *Favia*, the fractional absorption per coral area decreased with increasing colony area (Fig. 6). In all coral species, the larger colonies, with more total chlorophyll, absorbed more light. This absorption was positively correlated with colony area and also with areal chlorophyll concentration. Conversely, the percent absorbed photons per microgram chlorophyll decreased with increasing chlorophyll concentration (Fig. 6). Such a relationship was found in four of the five coral species that were examined: *Acropora* sp., *S. pistillata*, *P. damicornis*, *Favia* sp., and in the hydrocoral *M. dichotoma*, but not for *Fungia* sp. (Fig. 6)

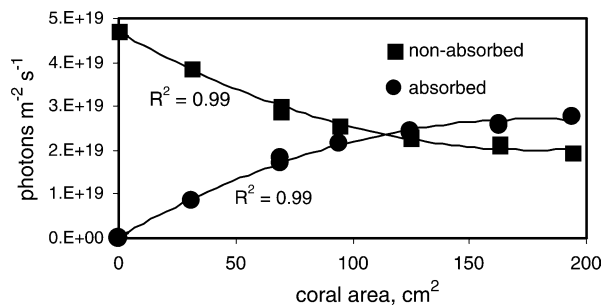


Fig. 3 The amount of light measured in the sphere and the light absorbed by the coral *Acropora* sp. as function of coral area ($n = 13$, polynomial regression $r^2 = 0.99$)

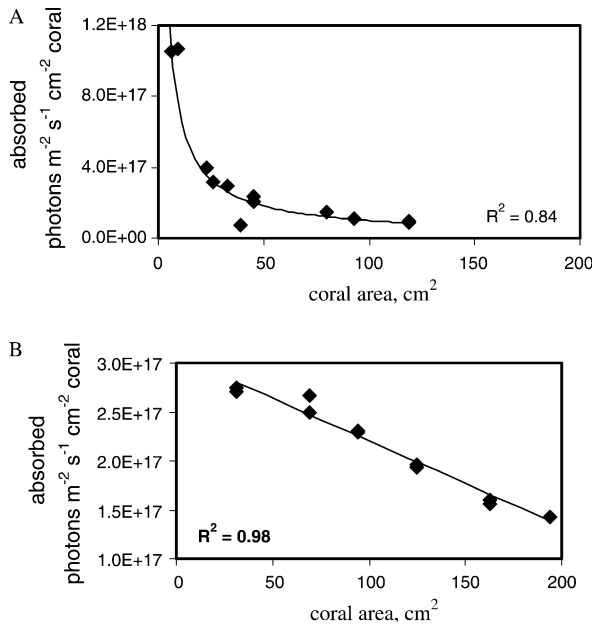


Fig. 4 The amount of light absorbed per surface area as a function of the coral size. (a) *Acropora* ($n=13$, power regression $y = 5 \times 10^{18} x^{-0.8585}$, $r^2 = 0.84$). (b) *Millepora* ($n=12$, linear regression $y = -8.7 \times 10^{14} + 3 \times 10^{17} x$, $r^2 = 0.98$)

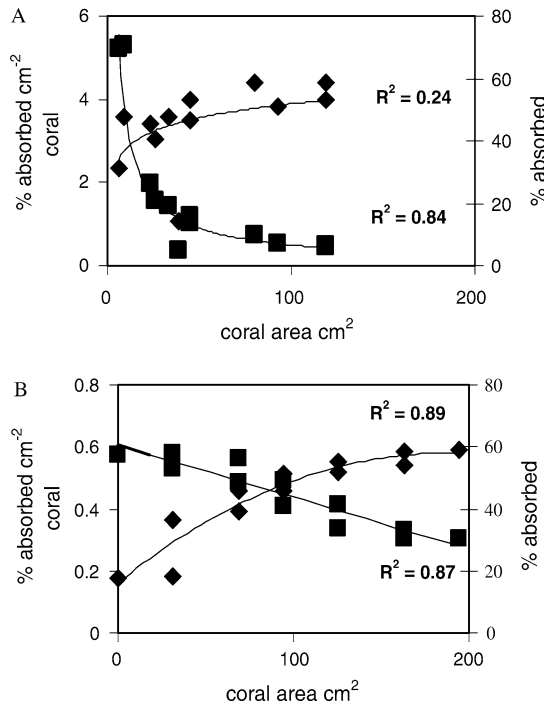


Fig. 5 (a) The fraction of light absorbed by *Acropora* coral colony (filled square), ($n=12$, logarithmic regression, $y = 6.3 \ln x + 22.6$, $r^2 = 0.84$), and per unit of colony area (filled diamond) ($n=12$, power regression, $y = 25.9x^{-0.86}$, $r^2 = 0.24$), as a function of coral area. (b) The fraction of light absorbed by *Millepora* coral colony (filled square), ($n=13$, polynomial regression, $r^2 = 0.87$), and per unit of colony area (filled diamond) ($n=13$, linear regression, $y = -0.002x + 0.6$, $r^2 = 0.89$), as a function of coral area

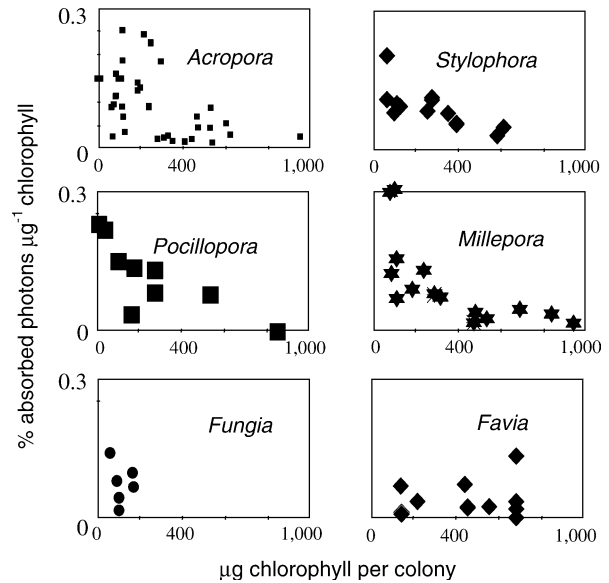


Fig. 6 Absorbed photons (%) per microgram chlorophyll as a function of chlorophyll content. Each graph represents different colonies of the same species

in which all samples had similar chlorophyll concentrations. There was high variability between the colonies of each species and no significant correlation was found.

The in vivo, chlorophyll-*a* specific, spectrally averaged, effective optical cross section, a^* , calculated from the integrating sphere, decreased with increasing areal chlorophyll concentration (Fig. 7, $r^2 = 0.57$, Table 1 ANOVA $p > 0.005$). High-light-adapted corals with lower chlorophyll per area had higher a^* values than their shade-adapted counterparts (Table 1). This was found by both integrating sphere and filter methods. It is noteworthy that the values obtained by the filter method were two to four times lower than those of the integrating sphere (Table 1).

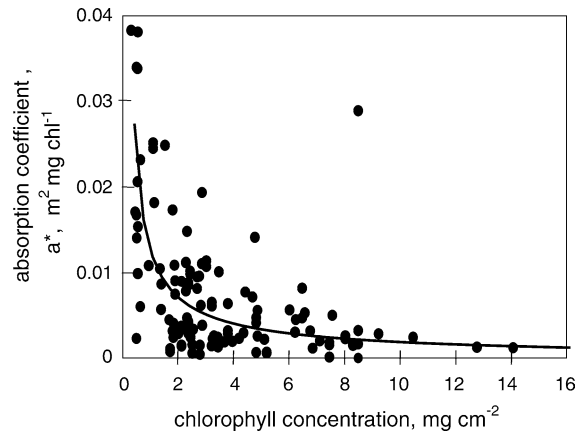


Fig. 7 The in vivo, chlorophyll-*a* specific, spectral average, effective optical cross-section, a^* , calculated from the integrating sphere, as function of chlorophyll concentration per coral surface area ($n=143$, power regression, $r^2 = 0.57$, $b = -0.725$, $a = 0.016$, ANOVA > 0.005)

Table 1 The in vivo, chlorophyll-*a* specific, spectral average, effective optical cross section, a^* , for *Stylophora pistillata* light and shade adapted corals. Data calculated from integrating sphere and

filters methods. The letters A, B, C, and D represent the results of the statistic tests ANOVA. Treatment with different letter are significantly different ($p > 0.005$)

	Chlorophyll content ($\mu\text{g cm}^{-2}$)	a^* by filters (m^2 per mg of chlorophyll)	a^* by integrating sphere (m^2 per mg of chlorophyll)
Light coral (HL) ($n=9$)	2.111 ± 0.702	0.0108 ± 0.0054 (A)	0.0235 ± 0.0060 (C)
Shade coral (LL) ($n=10$)	4.567 ± 1.868	0.0038 ± 0.0024 (B)	0.0153 ± 0.0060 (D)

Discussion

Most of the light that falls on corals is absorbed by the photosynthetic pigments of the zooxanthellae: chlorophylls *a* and *c* and carotenoids, mainly peridinin. The white calcium carbonate skeleton reflects light, as pointed out by Dustan (1982), thereby increasing the effective path length of light, and allowing light which was not absorbed by the zooxanthellae in the “first pass” to be captured by the zooxanthellae and the endolithic algae (Falkowski et al. 1990; Shashar and Stambler 1992; Kuhl et al. 1995; de Beer et al. 2000).

We found that the total amount of light absorbed by a colony always increased with colony size (Figs. 3–5). In spherical and planar colonies, the increase approximated a linear function of their area. However, this was not the case with branched colonies, because of the concomitant increase in self-shading as the colony becomes larger and morphologically more complex. Because of that very same geometric reason, the percent of light absorbed per coral area is higher for flat corals than for branched ones. Another way to visualize this idea is by pointing out that while the projected areas of coral colonies of the same radii, of flat or encrusting, massive subspherical, and branched subspherical will be the same, they differ substantially in their tissue and overall surface areas, the ratios being on the order of 1:~4:~10, respectively. Thus, for a given colony cross-section, or “footprint” the light harvested by a branched colony will be “diluted” over a far larger area than in spherical and flat forms.

The percentage of incident light absorbed by a unit of coral surface, in colonies of comparable morphology, would be expected to correlate with the product of the zooxanthellae areal density and the cellular pigment content. Theoretically, increasing the algal density would be expected to cause a corresponding increase in the absorption. This is true only as long as the algae are arranged in one layer. In nature, the algal density in several corals is around 1 million cells per square centimetre (Drew 1972) and, indeed, at such densities they may be accommodated in one layer (Kawaguti and Nakayama 1973). The maximal number of algae in 1 cm^2 of coral, assuming that the algae are spheres with a radius of $10 \mu\text{m}$, is ~ 1.5 million. In the case of *S. pistillata* in the Red Sea, zooxanthellae densities increased to 2.9×10^6 cells cm^{-2} upon exposure to N + P enrichment (Falkowski and Dubinsky 1981, Dubinsky et al. 1990). Working with *M. annularis*, Dustan (1979)

found a depth-dependent decrease in zooxanthellae densities from $8.76 \times 10^6 \text{ cm}^{-2}$ at 1 m, to $2.65 \times 10^6 \text{ cm}^{-2}$ at 45 m. Such densities require a multilayered arrangement. Whenever algae are arranged in more than one layer, there is bound to be an increasing overlap of cross-sections and self-shading. In such instances, the total amount of light absorbed per unit coral area will increase, while the amount of light absorbed by each zooxanthellae will inevitably decrease, due to an inter-cellular “package effect” (Geider and Osborne 1987).

High areal chlorophyll concentrations always increase the total light absorption, even though these also lead to a decrease in the percent absorption per chlorophyll unit (Fig. 6). Macdowall (1983), based on integrating sphere measurements, reported similar results for higher plants.

In the process of acclimation to low light intensity chlorophyll concentration of corals increases (e.g., Falkowski and Dubinsky 1981; Dubinsky et al. 1984; Thieberger et al. 1995; Stambler 1998, Table 1). However, exceptions to that rule have also been reported. Dustan (1979, 1982) and Titlyanov et al. (2001) have shown cases where areal chlorophyll increases with depth, down to a limit where it decreases sharply, in some cases due to a decrease in symbiont density. Below some threshold light intensity, which in the case of *S. pistillata* is $\sim 0.5\%$, there is not enough light to sustain any zooxanthellae at all, and the coral parts in such deep shade are white and hardly absorb any light (Dubinsky and Jokiel 1994).

In some cases, the bathymetric distribution of chlorophyll was found to be bimodal (Dustan 1979, 1982), possibly indicating the presence of two distinct symbiont populations (Rowan et al. 1997; Baker 2003). Dark, low-light acclimated corals absorb more light than their high-light conspecifics because they have more chlorophyll, but the efficiency of absorption, per unit chlorophyll is lower than that of high-light-adapted corals (Dubinsky et al. 1990). After the coral becomes a “black body” by increasing its chlorophyll concentration and its absorption approaches 100%, there is no way to further increase absorptivity. Any increment in areal chlorophyll will only result in increased inter- and intra-cellular overlap and reduction of the effective cross sections of these pigment molecules a^* (Schanz et al. 1997). Dustan (1982) already pointed out: “surprisingly, there was no absolute correlation between whole-cell absorption and absolute pigment content per zooxanthellae cell”.

The in vivo spectral average chlorophyll *a* absorption cross section, a^* (Kiefer and Mitchell 1983; Morel and Bricaud 1981) represents the probability of light harvesting by the pigment. There is a significant difference between the in vivo and the in vitro absorption because of the packaging (sieve) effect (Duysens 1956; Kirk 1976; Latimer 1983; Osborne and Geider 1989). The self-shading of the thylakoids causes additional difference in the mean absorption of the molecules (Berner et al. 1989).

In this study, we compared a^* values calculated from our integrating sphere measurements, and the filter method (Table 1). In the integrating sphere, unlike on the filter, the algae were packaged, arranged, and embedded in the coral tissue in a spatial arrangement different from that on the filter. Because of that, there is a difference between the resulting a^* values obtained by the two methods (Table 1). The filter method, due to the absence of coral tissue, and to the tighter packaging of the cells tends to enhance the difference in a^* between the HL (Light Coral) and LL (Shade Coral) adapted corals, as well as artificially decrease their values, in comparison with those calculated from the intact coral. On the filter the algae become arranged in many layers, which cause shading of the bottom ones by those above them. This situation results in an apparent decrease in the calculated cross-section (for discussion see Berner et al. 1989 and Schanz et al. 1997).

The in vivo a^* , calculated using the integrating sphere, shows a negative correlation to the areal chlorophyll concentration (Fig. 7). This result corresponds with, and explains, earlier reports that the pigment-specific light harvesting efficiency of shade-adapted/acclimated corals is lower than that of high-light ones (Table 1, Dubinsky et al. 1984). Since the areal chlorophyll concentration in intact corals is the product of the zooxanthellae density and of their cellular pigment content, we were unable to differentiate between the effects of these distinct intra- and intercellular packaging effects. Our findings are in agreement with those found in a previous study using the filter method on corals (Wyman et al. 1987; Lesser et al. 2000), and free phytoplankton (Kiefer et al. 1979; Dubinsky et al. 1986). The partitioning of that effect into the fraction due to pigment increase, stemming from growth in thylakoid area was done by Lesser and Shick (1990) and Stambler (1998). The increase in cellular pigmentation of the zooxanthellae is accomplished via increase in the size of the PSU (sensu Emerson and Arnold 1932), as has been shown by Falkowski and Dubinsky (1981), or increase in PSU numbers as observed by Lesser and Shick (1989) and Iglesias-Prieto and Trench (1994, 1997). The excise of both strategies may be due to different population or clades.

Corals from deep water are obviously exposed to far less light than those growing in shallow water ($\times 200$). This difference is compensated in part by the increased absorptivity of shade acclimated corals, that may absorb as much as 92% of incident PAR compared to only 50%

absorbed by high light corals (Dubinsky et al. 1984, Fig. 5). These results confirm those of Dustan (1982) who also underscored that corals at 30 m absorbed as much as 95% of incident light, thereby functioning as near-perfect black bodies. The difference between high- and low-light acclimated colonies is less than that which would result from the difference in their chlorophyll concentration, since it is reduced by the increased “package effects” and the reduced values of a^* (Berner et al. 1989; Kirk 1994; Schanz et al. 1997).

It seems that the additional shading of the zooxanthellae by the animal host tissue, either coral or sea anemone, may in several cases be ignored (Dubinsky 1980; Muller-Parker 1984; Hatcher 1988; Kuhl et al. 1995). This is probably not true for deeply pigmented hosts, like pink morphs of *S. pistillata* and *P. damicornis*, and blue *Acropora* species. One wonders if the bright tissue colours of many zooxanthellate corals, like *Dendrophyllia* sp., *Balanophyllia* sp and *Dendronephthia* sp. preclude the possibility of symbiosis.

Spherical corals such as *Favia* sp. and *Favites* sp. can increase their size and, as a result, the colony as a whole, will absorb more light (a), with no change whatsoever in flux per unit of living tissue. There may be an increase in chlorophyll concentration per alga (Table 1, Dubinsky et al. 1984) or an increase in algal density (Titlyanov 1991; Dubinsky and Stambler 1996), or a combination thereof. Both latter options are self-limiting due to the packaging effects discussed above (Dubinsky and Jokiel 1994). It is noteworthy that additional mechanisms of photoacclimation to low light were identified by Dustan (1975; 1982), Titlyanov (1991) and Titlyanov et al. (2001). Among the animal responses, a depth related decrease in polyp density in *M. annularis* (Porter et al. 1984a), may change the absorption/reflection fraction.

Branching corals such as *S. pistillata* and *Acropora* sp. have similar “architectural” options, but unlike in spherical and encrusting species, increasing colony size will enhance self-shading among branches. To avoid that, such corals change their shape to a more flattened morphology (Fricke and Schuhmacher 1983; Dubinsky and Jokiel 1994). In these species flat, platelike, horizontal growth was seen at the shaded base of the same colony, whereas brightly illuminated upper parts of the colony were profusely branched. The flattening response to low light also takes place in massive subspherical species. In both, the case of branched corals and that of massive ones, the flattening reduces the amount of tissue that has to be sustained by a given light flux, by reducing the ratio of surface (= tissue) to projected area. That was already pointed out by Goreau and Goreau (1959), Dustan (1975, 1979, 1982) and Roos (1967). The advantage of changing growth morphology according to light intensity is clearly demonstrated by the integrating sphere results (Figs. 2–6)

Characteristic coral reflection and fluorescence has been recently applied to discriminate between live coral cover and other benthos communities and follow coral bleaching episodes (e.g., Myers et al. 1999, Hochberg

et al. 2003, 2004). We think that information on coral absorption is important and that the non-disturbing integrating sphere method is a good technique, giving data on an entire colony.

The optical properties of zooxanthellate coral colonies are modulated in response to their light environment. Their responses combine changes in colony morphology, zooxanthellae density and algal pigment concentration. The integrating sphere method allowed us to examine some of these effects in intact colonies.

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