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Abstract. The present description of *Trichodesmium* spp. population dynamics and associated environmental variables is the first one using a very short sampling interval (2-3 days). Such a strategy allows a fine description of changes involving the population density and characteristics. It also lends itself to interpreting those changes from past environmental conditions. During the two surveys, 2 and 5 month-long, *T. erythraeum* dominated the *Trichodesmium* population, with temporary occurrence of *T. thiebautii* and five blooms could be described. These events appeared at temperatures > 24°C and followed, in all cases, nitrate, soluble reactive phosphorus and chlorophyll *a* enrichments, with a 3-7 day time lag. Low wind speed (< 4m s^{-1}) was not a prerequisite for *Trichodesmium* bloom developments as long as temperatures exceeded 26°C. As abundance increased during the bloom, so did the number of filaments (trichomes) in colonies and their buoyancy, leading to a clear positive biomass gradient from the bottom to the surface. A simple model, using variable growth rates, showed trichome ascent would be responsible for 87-99% of concentrations at 0.5m, with *Trichodesmium* net growth rates ranging from 0.11 to 0.38 d^{-1}. Finally, rapid trichome density declines could be ascribed to nutrient depletion and massive surface death following ascent.

1. Introduction

High surface concentrations of the chain-forming diazotrophic cyanobacteria *Trichodesmium* spp. have drawn attention, not only for their spectacular characteristics, but also for their potential toxicity (Landsberg, 2002,) and contribution to the carbon and nitrogen cycles in oligotrophic regions via diazotrophy, i.e. di-nitrogen (N_2) fixation (Karl et al., 2002). Such natural phenomena, called "blooms" in most references, have often been observed in the south-west Pacific, both in the open ocean (Dupouy et al., 2000) and coastal areas (Jones et al., 1982), as well as in other parts of the tropical belt (Capone et al., 1997). While many causative mechanisms for bloom development

have been proposed in the past (Sellner, 1997), we still lack reliable descriptions of past environmental conditions that triggered their increase. The word "bloom" itself remains imprecise and is generally understood as corresponding to rapid and intense growth, as it does for other phytoplankters, although it is now admitted that aggregation may be responsible for many observed surface *Trichodesmium* concentrations. One of the reasons for such uncertainties is that many reports are based on instant observations, no account being provided on past characteristics, biomass and vertical distribution of the *Trichodesmium* population, as stressed by Carpenter et al. (2004). Therefore, the understanding of bloom dynamics and causes would benefit from high frequency observations of both cyanobacteria and environmental variables over the water column.

Such a strategy, involving very frequent sampling on a long period of time can virtually be made in coastal areas, only. Present knowledge, however, rely on rather low frequency observations with sampling being made every 7 – 15 days (Lugomela et al., 2002), 1 month (Post et al., 2002) or 10 – 15 days (Muslim and Jones, 2003). While correlations between simultaneous data on *Trichodesmium* densities and environmental variables could be evidenced in these studies (Post et al., 2002; Muslim and Jones, 2003), effect of potential bloom triggers and environmental conditions that are required for bloom developments were not clearly proved because no information was collected in the few days preceding the start of the blooming event. For the same reason, causes of high surface aggregations remained hypothetical for lack of high temporal resolution surveys of *Trichodesmium* density along the water column, so that discriminating between effects of growth, vertical migrations and physical aggregation was impossible. Such considerations led to organizing “high” frequency sampling at the entrance of the Bay of Ste Marie (Noumea, New Caledonia) with two surveys made during the austral spring and summer, as both seasons are reported to be favourable for bloom development. Surveys lasted for 2.5 (Survey I: 9 October - 19 December 2003) and 5 months (Survey II: 15 November 2004 - 12 April 2005) with field measurements made at the same station, three times per week.

Here, we describe the *Trichodesmium* population, the changes in filament (trichome) density, degree of aggregation in colonies and vertical distribution during bloom development. From such a description and observed biovolume increase rates, we will show that high surface concentrations are due to growth and trichome ascent. Further, potential bloom triggers and favourable environmental conditions, will be identified from monitored environmental variables that prevailed between 2 and 7 days before. These variables were selected, taking into account results of the Muslim and Jones (2003) study in a similar shallow coastal site of the Great Barrier Reef (GBR, Australia): meteorology (wind strength and rainfall), hydrology (temperature and salinity), dissolved inorganic nutrients, turbidity, a proxy of suspended sediments, and chlorophyll, in addition to phytoplankton and zooplankton composition. Finally, one of the usual criticism of conclusions based on a solitary station deals with the effect of spatial heterogeneity on time-series data. This point will be considered taking into account observations made simultaneously at the entrance and 2.0 km off the bay.

2. Materials and methods

2.1. Sampling sites and general conditions (Fig. 1)

The study site, Sta. SM (22°18.86' S - 166°27.95' E, Fig. 1), was visited every Monday, Wednesday and Friday, between 07:00 and 09:00. While the station is away from inhabited areas and from urban inputs, it is protected from the prevailing SE trade winds and swell. A second station, Sta. O (22°19.80’ S – 166°28.48’ E) was visited in 2004-2005, weather permitting for comparison. Water column depth is 12 m and 28 m for Stas SM and O, respectively.

2.2. Meteorology, hydrology, and water sampling

Meteorological data were recorded at Faubourg Blanchot Meteo-France Station, 4.2 km away from the entrance of the Bay of Ste Marie.

CTD profiles (1 m to bottom) were obtained with a SeaBird® SBE 19 probe, fitted with pressure, temperature, salinity, turbidity, fluorescence and Photosynthetic Available Radiation (PAR) sensors.

Water samples were collected with 5 L-Niskin bottles at 0.5, 4, 7, and 10 m in 2003 and 0.5, 3, 6, and 10 m in 2004-2005. Niskin bottles were siphoned in the laboratory on land for nutrient and pigment analyses and *Trichodesmium* abundance, within one hour of sampling.

2.3. Nutrients and pigments

Nutrients were analyzed on $HgCl_2$-preserved samples from 0.5 and 6-7 m. Nitrate (NO_3) concentrations were determined by colorimetry using a Technicon® auto-analyzer and standard techniques (Strickland and Parsons, 1972), except for $NO_3 < 0.1 \mu M$, when the "high sensitivity" procedure of Raimbault et al. (1990) was used. Soluble reactive phosphorus (SRP) concentrations were measured with a Cecil® CE 1011 (10cm length-cell) spectrophotometer, using the molybdenum blue reaction (Murphy and Riley, 1962).

Chlorophyll *a* (Chl *a*) was measured on the < 20 and > 20 µm size fractions at all sampled depths, except for the > 20 µm fraction in 2004-2005, when analyses were only made at 3 and 10 m. For the < 20 µm Chl *a*, 200ml were prefiltered through a 20 µm nylon screen and collected on Whatman® GF/F filters. For the > 20 µm fraction, the whole Niskin bottle content was filtered through a 20 µm screen and gently rinsed off with filtered seawater onto GF/F filters. All filters were stored in liquid nitrogen until analysis within less than 2 weeks. Chl *a* was extracted in methanol and measured by fluorometry as described by Le Bouteiller et al. (1992).

2.4. Trichodesmium abundance and biometry

Trichodesmium were collected on 20 µm nylon screen from the total content of Niskin bottles (mean volume: 5.74 L) and transferred to a "sedimentation cell" with 5% formaldehyde. When trichomes did not sink, a few drops of pure acetic acid were added to the cell, in order to lower solution p_H and break gas vesicles (Carpenter and Carmichael, 1995). Microscopic examinations and

104 enumeration were made 24 h later using a Leitz® Fluovert inverted microscope. Enumeration
105 considered free trichomes and fusiform (tuft) and spherical (puff) aggregates, usually called
106 “colonies” in the literature. The number of trichomes per colony was recorded and the volume of
107 each trichome was calculated from its measured length and diameter, assuming a cylindrical shape
108 for the organism. In this paper, *Trichodesmium* abundance has been expressed in terms of total
109 number of trichomes (i.e. free and in colonies) per litre (trich. L^{-1}), or as trichome biovolume per
110 litre ($\mu m^3 L^{-1}$). Integrated values have been computed by the trapezoidal method. While trichome
111 density and biovolume are closely related (Fig. 2), due to the fact that biovolume data includes
112 trichome numbers, biovolume is often preferred as it gives a better estimate of the biomass.
113 Two species of *Trichodesmium* were identified according to Carpenter and Carmichael (1995)
114 and Janson et al. (1995). Individual cells of *T. thiebautii* Gomont are more long than wide and the
115 trichome has a convex apex whereas *T. erythraeum* Ehrenberg cells are more wide than long and the
116 trichomes have a flat apex. *T. erythraeum* colonies are usually darker than *T. thiebautii* colonies
117 (Carpenter et al., 1993). Such morphological differences between the two species have also been
118 confirmed by genetic finger prints of *Trichodesmium* spp. sampled in different areas of New
119 Caledonia, considering the hetR and rrs encoding for 16S rDNA genes, the internal transcribed
120 spacer (ITS) of the 16S-23S rDNA region and a highly iterated palindrome, HIP1 (Trottet, 2003).
121
122 *2.5. Trichodesmium C, N and chlorophyll a contents*
123 Plankton nets (35-µm mesh), towed at 1 m below the surface, were the source of 18-75 colonies
124 hand-picked, using a plastic inoculation loop, and transferred onto 25 mm Whatman GF/F filters for
125 Chl *a* measurements, or on pre-combusted filters for carbon (C) and nitrogen (N) analyses.
126 Filters for Chl *a* measurements were stored in liquid nitrogen and analyzed as described above.
127 For C, N determinations, filters were dried at 60°C and kept in a dessicator until analysis with a
128 Integra-CN PDZ EUROPA mass spectrometer. Single colonies were also selected for carbon

assimilation and nitrogen fixation experiments using ^{13}C and ^{15}N techniques (data not shown here, but reported in Le Borgne et al., 2006).

Carbon, N and Chl *a* contents are expressed per colony (col^{-1}) and per trichome ($trich.^{-1}$). Since analyses were made on colonies originating from plankton nets, the mean number of trichomes per colony was obtained from microscopic observations of aliquots originating from the same catches and processed as described before.

2.6. Microphytoplankton (>20µm) and Macrosetella abundance

Counts of the major phytoplankton taxa were conducted on samples from 2004-2005 survey. Selected taxa were diatoms, dinoflagellates and *Trichodesmium* spp. The two specific *Trichodesmium* spp. grazers, copepods *Macro-* and *Microsetella* spp. (Roman, 1978; O'Neil and Roman, 1994) were all enumerated on all samples. They were grouped into the "*Macrosetella*" taxon, since *Macrosetella gracilis* is the most abundant species in both our samples and lagoons of the west coast of New Caledonia (Binet, 1984).

2.7. Trichodesmium biovolume net increase and carbon-specific growth rates

Using observed *Trichodesmium* biovolume increases, ΔVi, exponential net increase rates, ki (in d^{-1}), can be calculated at depth, zi:

$$\Delta Vi = V_{f,i} - V_{o,i} \text{ with } V_{f,i} = V_{o,i}\, e^{ki\ t} \Rightarrow ki = \ln(V_{f,i}/V_{o,i})/(t_f - t_o) \quad (1)$$

where $V_{f,i}$ is the maximum biovolume of a peak period at time t_f and $V_{o,i}$, at the initial time t_o. Biovolume-based doubling time, $d'_{A,i}$ (in d), is equal to:

$$d'_{A,i} = \ln(2)/ki = \ln(2)\ (t_f - t_o)/(\ln(V_{f,i}/V_{o,i})) \quad (2)$$

155 The carbon-specific growth rate for 0.5m depth, $g_{C,0.5}$, was obtained from ^{13}C primary
156 productivity and elemental composition measurements, made on colonies in 2004-2005 (data
157 presented in Le Borgne et al., 2006). Calculations of $g_{C,0.5}$ used the following equation:
158
159 $$gc_{,0.5} = \ln((Co+\rho_C)/Co) \quad (3)$$
160
161 where Co is the initial carbon concentration, ρ_C is the daily rate of carbon fixation, and time is
162 understood as one day.
163
164 *2.8. Data processing*
165 Non-parametric and Gaussian statistics were used, following Snedecor and Cochran (1967).
166 Bravais-Pearson and Spearman's rank correlation coefficients are *r* and r_S, respectively, with level of
167 significance shown by * and ** for $p<0.05$ and $p<0.01$ significance, respectively; *n* is the number of
168 data per series, *s*, the standard deviation and *p*, the probability of rejection in the Wilcoxon and sign
169 tests.
170
171 **3. Results**
172
173 *3.1. Trichodesmium population dynamics: abundance, vertical distribution and net increase rates*
174 *Trichodesmium* spp. were always present, at least at one of the sampled depths and made almost
175 the totality of the pelagic filamentous cyanobacteria, *Katagnymene* spp. being quasi-absent. *T.*
176 *erythraeum* was the dominant species in 2003 (99.0% of the 5,513 counts) and 2004-2005 (83% of
177 the 10,650 counts) with *T. thiebautii* making the remainder.
178 Amidst a background of low trichome concentration, several periods of abrupt increase and
179 decline were observed during the two surveys (Fig. 3 and 4). When the abundance exceeded 5
180 trich.L^{-1} for at least two consecutive sampling times (i.e. 2-3 days), trichomes were considered as

thriving. Given the sharp variations, the term “bloom” will be used hereafter even though it is not quite appropriate, in the sense that the *Trichodesmium* never dominated the microphytoplankton, contributing a maximum of 27.7% of microphytoplankton counts.

Five blooms were observed with various features summarized in Table 1. Each bloom was characterized by one peak abundance, except in 2003, when the 3 consecutive maxima were interpreted as belonging to the same bloom (see Discussion). Peak amplitudes were variable from one event to another and that of February 2005, although low, was considered as a bloom because trichome densities were above 5 L^{-1} for 3 weeks. *T. erythraeum* was responsible for most of the blooms, except in December 2004 and February 2005 when contribution of *T. thiebautii* was significant. A striking point was the vertical distribution during the blooms, with a clear positive gradient of trichome densities from deep to shallow depths (Fig. 3 and 4).

During the peak periods, the exponential net increase rate, ki, (equation 1), ranged from 0.19 to 1.51 d^{-1} at 6-7 and 10m and from 0.52 to 3.29 d^{-1} at 3-4 and 0.5m (Table 2). These ki values were higher than carbon-specific growth rates, $g_{C, 0.5}$,calculated from ^{13}C uptake rates at 0.5m: 0.12-0.17 d^{-1} (Le Borgne et al., 2006). Exponential net increase rate calculations from biovolume variations were not possible outside the peak periods, while carbon-specific growth rates, $g_{C,0.5}$, provided by the ^{13}C uptake method were low, 0.022 d^{-1} on average (range: 0.017 – 0.050 d^{-1}).

Finally, density variations were synchronized and of the same magnitude at stations SM and O (Fig. 4) except from February 2005 on, when higher *Trichodesmium* densities were often found at the latter site. Growth rates, estimated from the ^{13}C experiments at both stations were similar (Le Borgne et al., 2006).

3.2. Trichodesmium population features: trichome size, degree of aggregation and chemical constituents

For both surveys, biovolume per trichome displayed quite a large range (Table 3), their length variability was greater than the width one and *T. thiebautii* trichomes were significantly longer than

207 those of *T. erythraeum* ($p < 0.001$). There was no significant size difference between trichomes from
208 0.5 and 6 m, as well as from Sta. SM and O (not shown) and between 2003 and 2004-2005 for both
209 species.

210 During the two surveys, *Trichodesmium* population consisted mostly of free trichomes (65 ± 22
211 and 63 ± 15 % in 2003 and 2004-2005, respectively) and colonies in tuft morphology prevailed
212 (98% of the total number of colonies). Tufts were typically small, made of 2 to 34 trich. col^{-1} while
213 puffs of *T. thiebautii* were made up of 7 to 123 trich. col^{-1}. As the number of free trichomes
214 increased during the peak periods, so did the number of trichomes in colonies, leading to a
215 significant correlation between the two forms ($p<0.01$; Fig. 5). Regression curves between densities
216 of trichomes in colonies and free trichomes were calculated for each of the two periods, using a
217 semi-log scale in order to take the anormal distribution of the free trichomes into account (Fig. 5).

218 *Trichodesmium* carbon, nitrogen and Chl*a* content data, presented in Table 4, were quite variable,
219 likely due to different physiological or trophic states of filamentous cyanobacteria. Based on
220 trichome densities and Chl *a* content, *Trichodesmium* Chl *a* was estimated as accounting for no more
221 than 21% of total Chl *a*. Atomic C/N ratio averaged 6.2 (range: 4.6 - 7.4), a value close to the slope
222 (6.75) of the C versus N regression line ($r^2 = 0.96$, $n=10$) and to the Redfield ratio.

223 To sum up, present surveys evidenced a shallower vertical distribution, higher growth rates and
224 more trichome aggregation in colonies during *Trichodesmium* blooms. These are the result of
225 environmental changes that are now considered.

226

227 *3.3. General environmental conditions and causes of bloom developments* (Fig. 6 and 7)

228 Both surveys started in austral spring, a season characterized by settled SE trade winds (> 6 $m.s^{-1}$)
229 and low rainfall. Sea temperature increased gradually until the beginning of February (Survey II),
230 reaching 29°C. Amidst this increasing trend, windy periods induced lower sea temperature and
231 higher salinity in spring. In summer and autumn, however, effect of wind on salinity was the
232 opposite, wind being associated to heavy rainfalls brought by tropical depressions, Kerry being one

of them in January 2005. Their major effect was high rainfall on the mainland and resulting lower lagoon salinity (<35.00).

Nutrient concentration increases followed local rainfall, as recorded at Noumea Met station in most instances. When they did not, they could be due, either to regional precipitation, not necessarily recorded by coastal stations, or to wind events leading to resuspension of shallow sediments and associated benthic nutrients (Muslim and Jones, 2003). The latter interpretation might indeed explain the nitrate peaks of November 2003 (Fig. 6) and March 2005 (Fig. 7) and the peak of SRP of January 2005, that took place during windy periods with no rainfall.

These increases in nutrient concentrations were not necessarily followed by total Chl *a* peaks for reasons probably linked to vertical mixing or trace metal inhibitions, as discussed later for *Trichodesmium*. Microphytoplankton contributed 15.9% (s = 8.0; n = 119) to total Chl *a* (>20 µm + < 20 µm) in 2003 and 8.0% (s = 7.2; n = 130) in 2004-2005. Their main peak period occurred in February 2005, following January nutrient inputs (Fig. 7). Diatoms dominated the microphytoplankton, contributing 82% of the counts and dinoflagellates made most of the remainder (11%).

Trichodesmium sp. started thriving at a water temperature, ranging from 24.2 to 28.6°C (Fig. 6 and 7). The vertical temperature gradient was variable from one bloom event to the other (0.05 to 0.84°C) and, therefore, did not seem to affect *Trichodesmium* development. The same conclusion may be drawn for the vertical salinity gradient which ranged between 0.00 and 1.00.

A calm period (mean wind speed < 4 m s^{-1}) preceded development of *Trichodesmium* blooms, except in January 2005 (Fig. 7) when the bloom started and continued during a windy period (mean wind speed before the bloom commenced: 7.3 m s^{-1}). In this case, however, temperature was > 26°C. In the other instances, windy periods did prevent *Trichodesmium* growth, as observed in November-December 2003, for example. Higher turbidity was not systematically associated with wind speed and rather low turbidity was the rule most of the time. We conclude turbidity, as measured by the CTD sensor, is not a good descriptor of blooming conditions.

Trichodesmium always (5/5 cases) throve after nutrient (NO_3, SRP) and Chl *a* concentration increases, that occurred within 3-7 days before the peak abundance (Fig. 6 and 7). Since it happened in all 5 cases, its occurrence was significant (sign test at $p< 0.10$). In addition, amplitude of the peak abundance at 0.5 m was significantly correlated ($r_S = 0.879$*) with NO_3 concentration at the start of the bloom (Fig. 8), but not with SRP.

In order to test the nutrient and Chl *a* trigger hypothesis, we have considered whether all of their increases were followed by blooms. During the two surveys, there were only two exceptions (5-7 November 2003 and 10-14 January 2005) when nutrient enrichments did not lead to any development. In those two cases, the nutrient-Chl *a* increases were followed by windy periods (Fig. 6 and 7) which could, therefore, prevent bloom development. However, as seen previously, the wind factor alone may not explain the second exception (January 2005) since temperature was >26°C. A lower than usual salinity may be the direct or indirect reason for the lack of *Trichodesmium* bloom as it was also for the lack of microphytoplankton biomass increase at the same period (Fig. 7).

In summary, according to the present observations, development of *Trichodesmium* blooms would follow nutrient and Chl *a* concentration increases with a 3-7 day lag, provided temperature is >26°C and no heavy and sustained rainfall occurs. The same conditions apply to > 24°C temperatures, as long as wind speed is low (< 4 m s^{-1}).

4. Discussion

4.1. Sampling strategy: are conclusions drawn from one station valid?

Study of *Trichodesmium* population dynamics employed high frequency sampling at Sta. SM, assuming the same watermass and planktonic population were sampled for a long enough period. This implies a rather long residence time, which is supported by the study of Jouon et al. (2006). Thus, the "e-flushing time", a proxy of the water residence time, is 15-25 days at the entrance of the bay, for a ~8 m s^{-1} (15.5 knots) SE wind velocity, and longer, of course, for lower wind speeds.

Besides, no significant tide effect was evidenced, especially in terms of salinity at 1m depth, because water exchanges proceed from both the south and the east of Sta. SM (Fernandez et al., 2006). Note also that *Trichodesmium* density and environmental parameter variations at Sta. SM displayed no erratic trends, at least at a period of a week. It appears, therefore, that this station lent itself to a satisfactory temporal description.

Moreover, present observations were representative of the surrounding lagoon waters, as seen for the following parameters: (1) *Trichodesmium* spp. densities and microplankton abundance at 0.5 m were in a fair agreement at the two stations even though *Trichodesmium* densities became higher out of the bay from February 2005 on. (2) Proportions of the two *Trichodesmium* species or of other microphytoplankton taxa were not significantly different at Sta. SM and O and (3) *Trichodesmium* spp. growth rates, as provided by the ^{13}C method, were similar at the two stations. All these arguments support the view of a reasonable spatial homogeneity in the sampled area, which is the prerequisite to temporal variation descriptions.

4.2. Trichodesmium population characteristics

Density as compared to other studies. Trichomes have been observed systematically during the two surveys and appeared to be a component of the usual phytoplankton population, as happens in the GBR where they would occur in 83% of the samples (Jones, 1992). However, they never made the bulk of the phytoplankton at Sta. SM, with their Chl *a* accounting for no more than 21% of the total. Similar contributions were reported by Letelier and Karl (1996) and Dupouy et al. (2000) for the open ocean of the tropical Pacific although higher values (11- 62%) were found in the Atlantic by Carpenter et al. (2004).

Maximum *Trichodesmium* abundances (240 and 244 trich. L^{-1} in 2003 and 2004, respectively) reported in this study, are rather modest compared to those found in the literature (Table 5) and this may be explained by differences in both methods and environmental conditions. Use of a 20µm mesh, trichome transfer from the silk into the sedimentation cell, insufficient trichome sinking

(Lugomela et al., 2002) or use of a 5.74 L sampling bottle instead of a net (Chang, 2000) may be reasons linked to the methodology. In addition, present results refer to samples at 0.5m while some of the reported densities in the literature refer to the very near-surface (i.e. 0m), where much higher concentrations could be associated with discoloured waters and accumulations along fronts or Langmuir cells. It remains some of the high densities found in literature refer to subsurface maxima (e.g. 12-24 m of Carpenter et al., 2004) and not to the very top surface, leading us to look for environmental-related reasons. Thus, an element may limit growth of the filamentous cyanobacteria and it is probably not dissolved iron, given its very significant concentrations in the SW lagoon, ranging between 2 and 5 nM (unpublished data). These values are much above 1 nM, the concentration given by Sanudo-Wilhelmy et al. (2003) for areas of non Fe-limited diazotrophy in the Atlantic ocean. Phosphorus, however, could be the limiting element of primary production, considering that its two possible sources are phosphorus depleted: runoffs from lateritic soils of this part of New Caledonia (Tenorio *et al.,* 2005) and exchanges with the surrounding oligotrophic ocean (Van Den Broeck et al., 2004). This limitation could explain both the modest trichome density maxima and the low Chl *a* (< 1 mg m^{-3}) concentrations (Fig. 6 and 7). Lastly, the observed positive and significant correlation found between trichome and diatom densities (r_S = 0.395**; n = 41) would suggest no negative effect of the main component of the microphytoplankton on *Trichodesmium*. In conclusion, modest trichome density maxima of the present study might result from a combination of different factors, the main one being possibly phosphorus limitation.

Specific composition. T. erythraeum was the dominant species with temporary occurrence of *T. thiebautii*. The two species have distinct morphometric characteristics and they displayed much higher length variations at Sta. SM (Table 3) than those reported by Post et al. (2002) in the Gulf of Aqaba: 300 - 800 µm for *T. erythraeum* and 1000 - 2000 µm for *T. thiebautii*.

T. erythraeum dominance at Sta. SM, as in the GBR lagoon (Jones, 1992; Muslin and Jones, 2003) or the Tanzanian coast (Lugomela et al., 2002 ; Bryceson and Fay, 1981), the Mississipi plume (Eleuterius et al., 1981) or the Bresilian coasts (Satô et al., 1963) might lead to the conclusion

it would be found more often in coastal areas than *T. thiebautii*. This seems to be corroborated by observations made in the open ocean off the east coast of New Caledonia, where *T. erythraeum* makes only 20-25% of the *Trichodesmium* filaments, while *T. thiebautii* and *T. tenue* would make 40-45% and 30% of the total, respectively (Tenorio, 2006). Actually, each of the two species is able to prevail in the open ocean. It is *T. erythraeum* in the Coral Sea and *T. thiebautii* in the Caribbean Sea (O'Neil et al., 1996), *T. thiebautii* in the Kuroshio area (Saino and Hattori, 1980) and the Atlantic (Carpenter et al., 2004), but *T. erythaeum* in the north Indian Ocean (Capone et al., 1998). Therefore, it seems unlikely that dominance of the two species would rely on the "hemisphere" as suggested by Capone and Carpenter (1999), but rather on physiological and/or environmental conditions, such as those prevailing in rather closed coastal areas, that seem to be more appropriate to *T. erythraeum*. Such a feature may be explained by a lower diazotrophic capacity of this species (Carpenter et al., 1993), which would imply its preference for less oligotrophic areas than *T. thiebautii*, on the one hand, and by less resistance of its vacuoles to pressure, which means less adaptation to deep environments, on the other hand (Carpenter et al., 1993).

Contribution of trichomes in colonies to total. Most (98%) of the colonies were of the tuft type and made of < 20 trichomes, on average. Similar numbers were found for *T. erythraeum* by Bryceson and Fay (1981) and Capone et al. (1998) with respectively, 5.5-10 and 10-30 trich. col^{-1}. But Carpenter et al. (2004), Letelier and Karl (1996) and Post et al. (2002) reported numbers > 100 for both *T. erythraeum* and *T. thiebautii*.

Also variable in the literature is the proportion of the number of aggregated trichomes to total number of trichomes, or "bundleness" (Bryceson and Fay, 1981): ratios <20% were observed in the North Pacific (Saino and Hattori, 1980 ; Letelier and Karl, 1996) and in the Atlantic (Orcutt et al., 2001 ; Tyrell et al., 2003) while bundleness ranged from 25 to 90% off the Tanzanian coast (Bryceson and Fay, 1981) and in the tropical Atlantic (Carpenter et al., 2004). Such variability is discussed by the latter authors and could originate from damages to the colonies during the sampling and sorting processes, too small a sampled volume, and/or a "fundamental difference in the state of

363 trichomes" between the different regions. Further, Orcutt et al. (2001), Fu and Bell (2003b) and Bell
364 et al. (2005) suggest there would be more of free trichomes during the optimum growth phase, and
365 more aggregation during the non optimum phases. Effect of wind speed and related water turbulence
366 on bundleness is invoked by Bryceson and Fay (1981) and would be negative, although the present
367 work does not support such a view, with bundleness not lower than usual during the windy January
368 2005 peak. More simply, present observations (Fig. 5) suggest bundleness would follow encounter
369 probability law, i.e. the higher the trichome density, the higher the probability, thus agreeing with the
370 view of Carpenter et al. (2004) of density-related bundleness. But, whatever the factors involved in
371 colony formation, high bundleness seems to be typical of *Trichodesmium* blooms and responsible for
372 higher nitrogen fixation rates (Saino and Hattori 1980; Bryceson and Fay, 1981; Letelier and Karl,
373 1998; Capone, 2001).
374
375 *4.3. Trichodesmium bloom features: respective roles of growth and ascent*
376 A striking result in the present study and many other publications, is the intensity of
377 *Trichodesmium* density increases, which appear to be inversely related to depth (Table 2): the
378 shallower the level, the greater the increase. Moreover, doubling times d'_{A}, calculated on biovolume
379 increases at 0.5, 3 - 4 and 6-7 m (Table 2), are much shorter than those reported in the literature for
380 growth (Table 6), while $d'_{A,10}$ at 10 m are in good agreement. From these observations, we conclude
381 $d'_{A,10}$ mainly result from growth and $d'_{A,i}$ of shallower depths, result from growth plus another
382 process. The latter is very likely the trichome ascent due to their positive buoyancy, a process which
383 delivers them more or less rapidly to the surface as they grow and which would explain the observed
384 vertical density gradient from the bottom to the surface (Fig. 3 and 4). Inversely, this gradient cannot
385 be ascribed to trichome sinking because microscopic observations showed healthy trichomes at all
386 depths and no or few trichomes were retrieved in sediment traps of the 2003 survey (Le Borgne et
387 al., 2004).

388 *Trichodesmium* positive buoyancy is well known and *T. erythraeum* has been reported more
389 buoyant than *T. thiebautii* (Carpenter et al., 1993; Lugomela et al., 2002), which would explain the
390 modest increases of February 2005 at 0.5 m when *T. thiebautii* contribution was significant. Such
391 positive buoyancy, linked to the presence of gas vesicles (Capone et al., 1997), is different from that
392 governing daily up and down vertical motion, with diel variations of the carbohydrate to protein ratio
393 (Villareal and Carpenter, 2003).

394 In order to test whether trichome biovolume increases (ΔVi) observed during the bloom periods,
395 resulted from both their net growth (Gi) and ascent (Ai) from deeper levels and in order to estimate
396 the respective contributions of the two processes, we used a simple model. Provided spatial
397 heterogeneity is negligible, as compared to temporal variability (see 4.1.), observed ΔVi at depth zi,
398 between t_0 and t_f (see eqn (1)) may be ascribed to the sum of Gi and Ai:

399

400 $$\Delta Vi = V_{f,i} - V_{o,i} = Gi + Ai \leftrightarrow \Delta Vi - (Gi + Ai) = 0 \quad (4)$$

401

402 with $Gi = \sum_{t=to}^{tf} V_{o,i}\, e^{gi\, t}\, \Delta t$, gi, being the exponential net growth rate, Δt, the time interval.

403 Corresponding doubling time, $d_{A,i}$, is equal to: $d_{A,i} = \ln(2)/gi$. Now, assuming trichome ascent, Ai,
404 would proceed at the same pace as growth and gi be the same along the water column, Ai will be the
405 integral of Gi with depth: $Ai = \sum_{z=12}^{zi} Gi\ \Delta z = \sum_{t=to}^{tf} \sum_{z=12}^{zi} V_{o,i}\, e^{gi\, t}\, \Delta t\ \Delta z$, where Δz is the difference
406 between two sampled depths and 12 m, the Sta. SM depth.

407 Calculations of Ai and Gi have been made for zi = 0.5 m, with the following Δz: 12-10 m, 10-7
408 m, 7-4 m, 4-0.5 m in 2003 and 12-6 m and 6-0.5 m in 2004-2005. We solved equation (4), and
409 considered the realism of computed doubling times, $d_{A,\,0.5}$ (Table 7). Except for the December 2004
410 event, $d_{A,\,0.5}$ values (1.82 - 6.45 d) are within the range of the doubling times in the literature (Table
411 6) and include the December 2003 biovolume doubling time at 10m, $d'_{A,\,10}$ = 2.9 d (Table 2). The
412 December 2004 low $d_{A,\,0.5}$ value (0.53 d) may be ascribed to an underestimated initial biovolume,

413 $Vo_{,0.5}$, which is used in equation 4. Estimates of $A_{0.5}$ and $G_{0.5}$ show that most (87 to 99%) of
414 biovolume increase observed at 0.5m ($\Delta V_{0.5}$), would be due to $A_{0.5}$. Ascent contribution variability
415 might be linked to the mixing intensity of the water column, as illustrated by the windy January 2005
416 bloom, which had the lowest ascent contribution (Table 7).
417 In this type of calculation applied to peak periods, the growth rates (gi) varied from one peak
418 abundance to another ($g_{0.5}$ = 0.11 – 0.38 d^{-1}). They varied also between non bloom and bloom
419 periods, as illustrated by the carbon-specific growth rates, provided by ^{13}C uptake measurements.
420 Indeed, a ratio of 5.1 - 7.4 (see Results) between bloom and "normal" periods may be computed,
421 leading us to the next question: what is the origin of gi variations leading to bloom developments?
422
423 *4.4. Causes of bloom developments: favorable conditions and triggers*
424 These may be shared into necessary conditions which allow blooms to occur, and triggers that
425 effect gi. The latter have been identified as nutrient increases during the 3-7 preceding days for all
426 the blooms, which supports observations made by Carpenter and Price (1977), Bell et al. (1999),
427 Muslim and Jones (2003) regarding a phosphate effect and Lugomela et al. (2002) about nitrate-
428 related blooms. Because diazotrophy is phosphorus-dependent (Sanudo-Wilhelmy et al., 2001; Fu
429 and Bell, 2003a; Mills et al., 2004), the role played by SRP in bloom developments seems to be
430 obvious. Less straightforward, however, may look the NO_3 increase effect on diazotrophic
431 cyanobacteria growth. In fact, di-nitrogen fixation is usually low or absent when other nitrogenous
432 compounds are available since it is high energy demanding and repressed by NH_4 (Mulholland and
433 Capone, 2000; Karl et al., 2002). At Sta SM, very low $^{15}N_2$ fixation rates were indeed measured on
434 *Trichodesmium* colonies during the March-April 2005 bloom (Le Borgne et al., 2006), suggesting
435 most of their nitrogen needs were fulfilled by other compounds. Ammonium and very small
436 dissolved organic nitrogen (DON) molecules, which are released or produced by the microbial loop,
437 seem to be the best candidates, although such a statement cannot be proved for a lack of
438 measurements.

Then we get to the following scenario which fits observations at Sta SM. Observed Chl *a* increases are very quick responses to NO_3 inputs. Part of the Chl *a* increases may be due to diatoms, which have a higher NO_3 uptake capacity than most phytoplankters (Sarthou et al., 2005), and likely respond immediately to nutrient inputs and lead to higher concentrations of ammonium and small DON molecules through bacterial remineralization and microzooplankton grazing. These are taken up eventually by filamentous cyanobacteria with a 3-7 d time-lag between NO_3 increases and *Trichodesmium* bloom initiation. The direct relation between *Trichodesmium* and diatom densities reported before may therefore be interpreted in this way.

The nutrient-related scenario suffers a troubling exception: the most important nutrient increase of the two surveys, in January 2005, was followed nor by *Trichodesmium* nor by microphytoplankton development, while the whole area was invaded by waters of lower than usual salinity. The salinity may not be incriminated *per se*, considering maximum *Trichodesmium* growth occurs in the 30 - 37 psu range (Fu and Bell, 2003b; Bell et al., 2005). It may rather indicate important terrigenous inputs of suspended sediments and heavy metals and their possible inhibition of microphytoplankton (including *Trichodesmium*) growth. Inhibition could proceed through light limitation due to a heavy load of suspended particles. No significant increase in turbidity (data not shown) was observed, however. Different could be the heavy metals inhibitory effect and Fernandez et al. (2006) have shown Fe, Ni, Cr, Co, Mn, Cu and Sr are issued from laterite weathering, and carried along from several estuaries, south of Noumea (e.g., La coulee, Fig. 1). There may be also an impact of the amount and type of organic matter from terrigenous origin, on the microphytoplankton, although this topic has not been documented, yet.

Bloom triggers work, provided necessary conditions are met, *i.e.* appropriate iron concentration, temperature and wind velocity. As seen above, iron is probably non limiting in the present study, but sunny and calm weather conditions may be necessary ones as invoked in the past by many authors (Eleuterius et al., 1981; Jones et al., 1992; Sellner, 1997; Lugomela et al. (2002) ; Muslim and Jones, 2003 ; Carpenter et al., 2004). However, calm weather is not sufficient a condition because

there may be blooms during long periods of wind, as illustrated by the January 2005 event. Based on this example, when temperature was ~ 26°C, we conclude that blooms may occur during windy periods, only if sea temperature is above this threshold value, thus confirming the Carpenter and Capone (1992) analysis on bloom conditions. Inversely, below 26°C, a low wind velocity appears to be necessary for bloom development. The minimum temperature for them to occur is 24°C in the present study, which is less than the 25°C threshold of Chen et al. (2003). Combination of wind and temperature effects can be interpreted as the result of two antagonist actions on growth rates, positively correlated to temperature and negatively to wind induced-mixing. It follows that, when temperature reaches a threshold value, its effect on growth will overcome the negative effect of mixing.

4.5. Decline origin

One of the striking points in the two surveys, was the brevity of the bloom decline phase, as for the growing one. Decline may be ascribed to mortality and to a recruitment diminution due to a growth rate slowdown as for any population, even though they might have common origins. In the present study, most blooms ended with very low nutrient concentrations (<0.030 µM NO_3 at the end of the December 2003 and February 2005 events and <0.030 µM SRP for those of February and April 2005), which suggests nutrient exhaustion represents one of the decline causes, as mentioned already by Lenes et al. (2005) and Moutin et al. (2005). This process could reduce growth rates, in a way opposite to the nutrient increase effect described above, or induce mortality through a "Programmed Cell Death" (PCD) pathway (Berman-Frank et al., 2004, 2007). Viral lysis (Ohki, 1999) and PCD, caused by other factors than nutrient depletion, as listed by Berman-Franck et al. (2004), are other possible processes. Inversely, intense grazing is likely excluded, considering the low *Macrosetella* densities that were observed and their lack of relationship with trichome concentrations. But the present observations on trichome ascent and surface accumulation during bloom periods with no obvious sinking or downward migration do not exclude massive destruction

by solar radiation as infra-red or ultra-violet rays, independently of other processes. The solar effect is likely linked to the sea surface agitation, with a more rapid destruction during calm sea and low wind conditions. On the whole, apart from trichome viral lysis or PCD, abrupt decay could be the result of ascent to the very top surface, solar destruction and lack of new trichome formation for nutrient exhaustion.

4.6. Summary of Trichodesmium bloom dynamics as illustrated by the November - December 2003 event

A summary of present conclusions on the bloom triggering factors (nutrient concentration increases), necessary conditions (combination of wind and temperature effects) and causes of bloom decay can be illustrated by the Nov-Dec 2003 event. Although it was made of three peak abundances, the following description shows it was only one event (Fig. 6).

An increase in nutrient concentrations was observed on 19 - 21 November for NO_3 and 17 November for SRP. At that time, water temperature was around 24°C and development could not start before the wind velocity diminished. This occurred on the 23rd and was likely responsible for the observed trichome density and biovolume increase between 21 and 28 November. On 29 November, while temperature was now 25°C, but still < 26°C, the wind started blowing again, which resulted in the first trichome density diminution between 28 November and 3 December. The wind slowed down between 4 and 5 December, allowing another *Trichodesmium* increase with a peak on the 5th. Calm weather conditions lasted until the 11th, allowing trichome ascent and mortality at 0m with the resulting biovolume decrease at all depths between 5 and 8 December and a small development between 10 and 12 December, once temperature had reached 26°C. The bloom ended when NO_3 became exhausted (< 0.005 µM at 0.5m), i.e. from the 10th, on.

5. Conclusion

Photosynthetic filamentous cyanobacteria of the *Trichodesmium* genus, display the same physiological processes as the other co-occuring phytoplankton and have the same requirements. In particular, dissolved nutrients are primarily involved in their growth processes. But the present surveys have shown major differences, with very steep density variations and changes in the cyanobacterial vertical distribution during the blooms, both being the result of the trichome positive buoyancy. This characteristic has two consequences: (1) most of the biomass accumulation will happen at 0m and not in the water column; (2) once they have reached the very top surface, trichomes will remain there, and eventually be destroyed. In addition, because of their toxicity, the grazing control of the *Trichodesmium* population appears to be restricted to a few species, like harpacticoid copepods (Hawser et al., 1992) and these were not abundant in the studied area. These two features of the *Trichodesmium* population dynamics, i.e. effects of positive buoyancy and low grazing losses, make it quite different from what happens to the rest of the phytoplankton community. Such a statement, however, is based on observations of a *T. erythraeum* dominated population, which is known to be more buoyant and less diazotrophic than *T. thiebautii*. Therefore, bloom causes and characteristics of *T. thiebautii* populations might be different to some extent and would deserve a similar study, related to the toxicity issue, this species being considered as more toxic than *T. erythraeum* (Sellner, 1997; Landsberg, 2002).

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References

Bell, P.R.F., Elmetri, I., Uwins, P., 1999. Nitrogen fixation by *Trichodesmium spp*. in the Central and Northern Great Barrier Reef lagoon: relative importance of the fixed-nitrogen load. Mar. Ecol. Prog. Ser. 186, 119-126.

Bell, P.R.F., Uwins, P.J.R., Elmetri, I., Phillips J.A., Fu, F.X., Yago, A.J.E., 2005. Laboratory culture studies of Trichodesmium isolated from the Great Barrier Reef Lagoon, Australia. Hydrobiologia 532, 9-21.

Berman-Frank, I., Bidle, K. D., Haramaty, L., Falkowski, P., 2004. The demise of the marine cyanobacterium, *Trichodesmium spp.,* via an autocatalyzed cell death pathway. Limnol. Oceanogr. 49, 997-1005.

Berman-Frank, I., Rosenberg, G., Levitan, O., Haramaty, L., Mari, X., 2007. Coupling between autocatalytic cell death and transparent exopolymeric particle production in the marine cyanobacterium *Trichodesmium*. Environt. Microbiol. doi. : 10.1111/j. 1462-2920.2007.01257.x.

Binet, D., 1984. Copépodes planctoniques du lagon de Nouvelle-Calédonie: facteurs écologiques et associations d'espèces. Mar. Biol. 82**,** 143-156.

Bryceson, I., Fay, P., 1981. Nitrogen fixation in *Oscillatoria* (*Trichodesmium*) *erythraea* in relation to bundle formation and trichome differenciation. Mar. Biol. 61, 159-166.

Capone, D. G., Carpenter, E. J., 1999. Nitrogen fixation by marine cyanobacterium : historical and global perspectives. Bull. Inst. Océanogr. Monaco 19, 235-256.

Capone, D. G., Zehr, J. P., Paerl, H. W., Bergman, B., Carpenter, E. J., 1997. *Trichodesmium*, a globally significant marine cyanobacterium. Science 273, 1221-1229.

Capone, D. G., Subramaniam, A., Montoya., J. P., Voss, M., Humborg, C., Johansen, A. M., Siefert, R. L., Carpenter, E. J., 1998. An extensive bloom of N_2-fixing cyanobacterium *Trichodesmium erythraeum* in the central Arabian Sea. Mar. Ecol. Prog. Ser. 172, 281-292.

Carpenter, E. J., Price, C. C., 1977. Nitrogen fixation, distribution, and production of *Oscillatoria* (*Trichodesmium*) spp. in the western Sargasso and Caribbean Seas. Limnol. Oceanogr. 22, 60-72.

Carpenter, E.J., Romans, K., 1991. Major role of the cyanobacterium *Trichodesmium* in nutrient cycling in the North Atlantic ocean. Science 254, 1356-1358.

Carpenter, E. J., Capone, D. G., 1992. Nitrogen fixation in *Trichodesmium* blooms. In: Carpenter, E. J., Capone, D. G., Rueter, J. G. (Eds.), Marine Pelagic cyanobacteria: Trichodesmium and other Diazotrophs. Kluwer Academic Publishers, Dordrecht, pp. 211-217.

Carpenter, E. J., Carmichael, W. W., 1995. Taxonomy of Cyanobacteria,. In: Hallegraeff, G. M., Anderson, D. M., Cembella, A. D. (Eds), Manual on Harmful marine algae. UNESCO, Paris, pp. 373-380.

Carpenter, E. J., O'Neil, J. M., Dawson, R., Capone, D. G., Siddiqui, P. J. A., Roenneberg, T., Bergman, B., 1993. The tropical diazotrophic phytoplankter *Trichodesmium* : biological characteristics of two common species. Mar. Ecol. Prog. Ser. 95, 295-304.

Carpenter, E. J., Subramaniam, A., Capone, D. G., 2004. Biomass and primary productivity of the cyanobacterium *Trichodesmium spp*. in the tropical N Atlantic ocean. Deep-Sea Res. I 51, 173-203.

Chang, J., 2000. Precision of different methods used for estimating the abundance of the nitrogen-fixing marine cyanobacterium, *Trichodesmium* Ehrenberg. J. Exp. Mar. Biol. Ecol. 245, 215-224.

Chen, Y-B. L., Chen, H-Y., Lin, Y-H., 2003. Distribution and downward flux of *Trichodesmium* in the South China Sea as influenced by the transport from the Kuroshio Current. Mar. Ecol. Prog. Ser. 259, 47-57.

Dupouy, C., Neveux, J., Subramaniam, A., Mulholland, M. R., Montoya, J. P., Campbell, L., Carpenter, E. J., Capone, D. G., 2000. Satellite captures *Trichodesmium* blooms in the southwestern tropical Pacific. Eos 81, 15-16.

Eleuterius, L., Perry, H., Eleuterius, C., Warren, J., Caldwell, J., 1981. Causative analysis on a nearshore bloom of *Oscillatoria erythraea* (*Trichodesmium*) in the Northern Gulf of Mexico. Northeast Gulf Science 5, 1 -11.

Fernandez, J-M., Ouillon, S., Chevillon, C., Douillet, P., Fichez, R., Le Gendre, R., 2006. A combined modelling and geochemical study of the fate of terrigenous inputs from mixed natural and mining sources in a coral reff lagoon (New Caledonia). Mar. Pollut. Bull. 52, 320-331.

Fu, F.-X., Bell, P. R. F., 2003a. Factors affecting N_2 fixation by the cyanobacterium *Trichodesmium* sp. GBRTRLI101. FEMS Microbiol. Ecol. 45, 203-209.

Fu, F.-X., Bell, P. R. F., 2003b. Effect of salinity on growth, pigmentation, N_2 fixation and alkaline phosphatase activity of cultured *Trichodesmium* spp. Mar. Ecol. Prog. Ser. 257, 69-76.

Hawser, S.P., O'Neil, J.M., Roman, M.R. Codd, G.A., 1992. Toxicity of blooms of the cyanobacterium Trichodesmium to zooplankton. J. Applied Phycol. 4, 79-86.

Janson, S., Siddiqui, P. J. A., Walsby, A.E., Romans, K.M., Carpenter, E. J., Bergman, B., 1995. Cytomorphological characterization of the planktonic diazotrophic cyanobacteria *Trichodesmium spp*. from the Indian Ocean and Caribbean and Sargasso Seas. J. Phycol. 31, 463-477.

Jones, G. B., 1992. Effect of *Trichodesmium* blooms on water quality in the Great Barrier Reef Lagoon,. In: Carpenter, E. J., Capone, D. G., Rueter, J. G. (Eds.), Marine Pelagic Cyanobacteria: *Trichodesmium* and other Diazotrophs. Kluwer Academic Publishers, Dordrecht, pp. 273-287.

Jones, G.B., Burdon-Jones, C., Thomas, F.G., 1982. Influence of *Trichodesmium* red tides on trace metal cycling at a coastal station in the Great Barrier Reef Lagoon. Oceanol. Acta Special Publication, 319-326.

Jouon, A., Douillet, P., Ouillon, S., Fraunié, P., 2006. Calculations of hydrodynamic time parameters in a semi-opened coastal zone using a 3D hydrodynamic model. Cont. Shelf Res.26, 1395-1415.

Karl, D., Michaels,A., Bergman, B., Capone, D. G., Carpenter, E.J., Letelier, R., Lipschultz, F., Paerl, H., Sigman, D., Stal, L., 2002. Dinitrogen fixation in the world's oceans. Biogeochemistry 57/58, 47-98.

Landsberg, J.H., 2002. The effects of harmful algal blooms on aquatic organisms. *Rev. Fish. Sci.* 10, 113-390.

Le Borgne, R., Faure, V., Raimbault, P., Rodier, M., 2004. Suivi de l'abondance de *Trichodesmium spp*. (cyanobactéries filamenteuses) et des paramètres du milieu lagonaire en Baie de Sainte-Marie (Nouméa, Nouvelle-Calédonie):7 octobre - 19 décembre 2003. Arch. Sc. Mer IRD/Nouméa 6, 1-50.

Le Borgne, R., Mazzeo, I., Raimbault, P., Rodier, M., Rouchon, C., 2006. Suivi de *Trichodesmium spp*. (cyanobactéries filamenteuses) et des paramètres du milieu lagonaire en baie de Sainte-Marie, Nouméa, Nouvelle-Calédonie. 3 novembre 2004 - 12 avril 2005. Arch. Sc. Mer IRD/Nouméa 8, 1-55.

Le Bouteiller, A., Blanchot, J., Rodier, M., 1992. Size distribution patterns of phytoplankton in the western Pacific: towards a generalization for the tropical ocean. Deep-Sea Res 39, 803-823.

Lenes, J.M., Walsh, J.J., Otis, D.B., Carder, K.L., 2005. Iron fertilization of *Trichodesmium* off the west coast of Barbados: A one-dimensional numerical model. Deep-Sea Res. I 52, 1021-1041.

Letelier, R. M., Karl, D. M., 1996. Role of *Trichodesmium* spp. in the productivity of the subtropical North Pacific Ocean. Mar. Ecol. Prog. Ser. 133, 263-273.

Lugomela, C., Lyimo, T. J., Bryceson, I., Semesi, A. K., Bergman, B., 2002. *Trichodesmium* in coastal waters of Tanzania: diversity, seasonality, nitrogen and carbon fixation. Hydrobiologia 477, 1-13.

Mills, M. M., Ridame, C., Davey, M., La Roche, J., Geider, R. J., 2004. Iron and phosphorus co-limit nitrogen in the tropical North Atlantic. Nature 429, 292-294.

Moutin, T., Van Den Broeck, N., Beker, B., Dupouy, C., Rimmelin, P., Le Bouteiller, A., 2005. Phosphate availability controls *Trichodesmium* spp. biomass in the SW Pacific Ocean. Mar. Ecol. Prog. Ser. 297, 15-21.

Mulholland, M. R., Capone D. G., 2000. The nitrogen physiology of the marine N_2-fixing cyanobacterium *Trichodesmium* spp.. Trends Plant Sci. 5, 148-153.

Mulholland, M. R., Bernhardt, P. W., 2005. The effect of growth rate, phosphorus concentration, and temperature on N_2 fixation, carbon fixation, and nitrogen release in continuous cultures of *Trichodesmium* IMS 101. Limnol. Oceanogr. 50, 839-849.

Murphy, J., Riley, J.P., 1962. A modified single solution method for the determination of phosphate in natural waters. Anal. Chim. Acta 26, 31-36.

Muslim, I., Jones, G.B., 2003. The seasonal variation of dissolved nutrients, chlorophyll *a* and suspended sediments at Nelly Bay, Magnetic Island. Estuar. Coastal Shelf Sci. 57, 445-455.

Negri, A.P., Bunter, O., Brian, J., Llewellyn, L., 2004. Effects of the bloom-forming alga *Trichodesmium erythraeum* on the pearl oyster *Pinctada maxima*. Aquaculture 232, 91-102.

Ohki, K., 1999. A possible role of temperate phage in the regulation of *Trichodesmium* biomass. Bull. Inst. Océanogr. Monaco 19, 287-291.

O'Neil, J. M., Roman M. R., 1994. Ingestion of the cyanobacterium *Trichodesmium* spp. by pelagic harpacticoid copepods *Macrosetella*, *Miracia* and *Oculosetella*. Hydrobiologia 292/293, 235-240.

O'Neil, J.M., Metzler, P.M., Glibert, P.M., 1996. Ingestion of $^{15}N_2$-labeled *Trichodesmium spp*. and ammonium regeneration by the harpacticoid copepod *Macrosetella gracilis*. Mar. Biol. 125, 89-96.

Orcutt, K. M., Lipschultz, F., Gundersen, K., Arimoto, R., Michaels, A. F., Knap, A. H., Gallon, J. R., 2001. A seasonal study of the significance of N_2 fixation by *Trichodesmium* spp. at Bermuda Atlantic Time-series Study (BATS) site. Deep-Sea Res. II 48, 1583-1608.

Post, A. F., Dedej, Z., Gottlieb, R., Li, H., Thomas, D. N., El-Absawi, M., El-Naggar, A., El-Gharabawi, M., Sommer, U., 2002. Spatial and temporal distribution of *Trichodesmium* spp. in the stratified Gulf of Aqaba, Red Sea., Mar. Ecol. Prog. Ser. 239, 241-250.

Prufert-Bebout, L., Paerl, H.W., Lassen, C., 1993. Growth, nitrogen fixation and spectral attenuation in cultivated Trichodesmium species. Appl. Environ. Microbiol. 59, 1367-1375.

Raimbault, P., Slawyk, G., Coste, B., Fry, J., 1990. Feasibility of using an automated colorimetric procedure for the determination of seawater nitrate in the 0 to 100 nM range: examples from field and culture, Mar. Biol. 104, 375-351.

Revelante, N., Gilmartin, M., 1982. Dynamics of phytoplankton in the Great Barrier Reef Lagoon. J. Plankton Res. 4, 47-76.

Riedel, G.F., J. G. Sanders, Breitburg, D. L., 2003. Seasonal Variability in Response of Estuarine Phytoplankton Communities to Stress: Linkages between Toxic Trace Elements and Nutrient Enrichment. Estuaries 26, 323-338.

Roman, M.R., 1978. Ingestion of the blue-green algae *Trichodesmium* by the harpacticoid copepod, *Macrosetella gracilis*. Limnol. Oceanogr. 23, 1245-1255.

Saino, T., Hattori, A., 1980. Nitrogen fixation by Trichodesmium and its significance in nitrogen cycling in the Kuroshio area and adjacent waters. In: Takenouti, A.Y. (Ed), The Kuroshio. Saikon Publishing, Tokyo, pp. 697-709.

Sanudo-Wilhelmy, S. A., Kustka, A. B., Gobler, C. J., Hutchins, D. A., Yang, M., Lwiza, K., Burns, J., Capone, D. G., Raven, J. A., Carpenter, E. J., 2001. Phosphorus limitation of nitrogen fixation by Trichodesmium in the central Atlantic Ocean. Nature 411, 66-69.

Sarthou, G., Timmermans, K. R., Blain, S., Tréguer, P., 2005. Growth physiology and fate of diatoms in the ocean: a review. Journal of Sea Research 53, 25-42.

Satô, S., Paranagua, M.N., Eskinazi, E., 1963. On the mechanism of red tide of *Trichodesmium* in Recife, northeastern Brazil, with some considerations of the relation to the human disease "Tamandare fever". Trab. Inst. Oceanogr. Univ. Recife 6/7, 7-49.

Sellner, K.G., 1997. Physiology, ecology and toxic properties of marinecyanobacteria blooms. Limnol. Oceanogr 42, 1089-1104.

Snedecor, G.W., Cochran, W.G., 1967. Statistical methods, 6th ed.. Iowa State Univ. Press.

Strickland, J., Parsons, T., 1972. A practical handbook of seawater analysis, Fish Res. Bd Can. Bul. 167.

Tenório, M. M. B., Le Borgne, R., Rodier, M., Neveux, J., 2005. The impact of terrigeneous inputs on the Bay of Ouinné (New Caledonia) phytoplankton communities : a spectrofuorometric and microscopic approach. Estuar. Coastal Shelf Sci. 64, 531-545.

Tenório, M. M. B., 2006. Les cyanobactéries pélagiques en milieu tropical oligotrophe: occurrence, distribution et dynamique. Univ. Paris VI Thesis, 1-359.

Trottet, A., 2003. Biodiversité des cyanobactéries filamenteuses du genre *Trichodesmium* dans l'océan Pacifique tropical sud-ouest. Mémoire DEA Univ. Paris VI, 1-26.

Tyrell, T., Maranon, E., Poulton, A. J., Bowie, A. R., Harbour, D. S., Woodward, E. M. S., 2003. Large-scale latitudinal distribution of *Trichodesmium* spp. in the Atlantic Ocean. J. Plankton Res., 25, 405-416.

Van Den Broeck, N., Moutin, T., Rodier, M., Le Bouteiller, A., 2004. Seasonal variations of phosphate availability in the SW Pacific Ocean near New Caledonia. Mar. Ecol. Prog. Ser. 268, 1-12.

Villareal, T. A., Carpenter, E. J., 2003. Buoyancy regulation and potential for vertical migration in the oceanic cyanobacterium *Trichodesmium*. Microb. Ecol., 45, 1-10.

Tables

Table 1

Main characteristics of the five *Trichodesmium erythraeum* blooms (* denotes presence of *T. thiebautii*).

Period	Number of peaks	Difference in time evolution at 6 and 0.5 m	Total duration (d)	Maximum at 0.5 m (trich. L^{-1})	Maximum at 6-7 m (trich. L^{-1})
24 Nov - 14 Dec 2003	3	No	18	238.7	47.4
13 - 20 Dec 2004*	1	at 6m, 2 d before	7	244.6	20.7
3 - 14 Jan 2005	1	No	11	81.5	14.8
4 - 18 Feb 2005*		No	14	10.1	19.4
29 Mar - 7 Apr 2005	1	at 6m, 4 d before	9	156.5	44.3

Table 2

Exponential net increase rates (ki in d^{-1}) and corresponding doubling times ($d'_{A,i}$ in d) in parentheses, from changes in *Trichodesmium* population biovolume, at different depths (equations 1 and 2).

2003	*T. erythraeum*				2004-2005	*T. erythraeum*		*T. thiebautii*	
	10m	7m	4m	0.5m		6m	0.5m	6m	0.5m
24-30 Nov		0.36	0.43	0.36	13-12 Dec	0.39	3.29	1.51	2.20
		(1.9)	(1.6)	(1.9)		(1.8)	(0.2)	(0.5)	(0.3)
3-9 Dec	0.24	0.68	0.61	0.72	3-14 Jan	0.98	1.13		
	(2.9)	(1.0)	(1.1)	(1.0)		(0.7)	(0.6)		
11-14 Dec			0.15	0.40	4-18 Feb	0.19	0.52	0.23	0.94
			(4.6)	(1.7)		(3.7)	(1.3)	(2.9)	(0.7)
					29 Mar - 4 Apr	0.67	0.59		
						(1.0)	(1.2)		

Table 3

Sizes of *T. erythraeum* and *T. thiebautii* during the 2003 and 2004-2005 surveys at Sta SM. Means ± standard deviations, and range (in parenthesis) of size parameters for *n* observations.

		Width (µm)	Length (µm)	Volume (10^3 µm^3)
T. erythraeum				
2003	0.5m - 10 m	11.0 ± 0.6	477 ± 190	45.3 ± 20.7
	n= 2612	(5.5 - 22.0)	(56-1489)	(5.3-426.1)
2004-2005	0.5m - 6 m	10.5 ± 1.4	550 ± 344	48.4± 34.8
	n= 6362	(5.5 - 22.0)	(60 – 2675)	(4.1 - 448.6)
T. thiebautii				
2003	0.5m – 10m	11.3 ± 2.3	822 ± 499	67.1 ± 26.0
	n=8	(8.2 - 16.5)	(440 -2031)	(25.6 - 96.9)
2004-2005	0.5m – 6m	5.7 ± 1.0	847 ± 440	22.0 ± 16.4
	n = 1618	(5.5 - 11.0)	(57 – 3386)	(1.3 - 186.4)

Table 4

Trichodesmium carbon, nitrogen and chlorophyll *a* content of the 2004-2005 survey: means, standard deviations (sd) and ranges.

	Per colony			Per trichome			Ratio	
	Chl *a* (ng)	C (µg)	N (µg)	Chl *a* (ng)	C (ng)	N (ng)	C/N (mol/mol)	C/Chl *a* (g/g)
Mean ± sd	7.84 ± 5.16	1.25 ± 0.29	0.20 ± 0.12	0.44 ± 0.26	103.1 ± 36.1	16.8 ± 5.7	6.2 ± 0.8	265.1 ± 161.5
Range	2.41 – 14.9	0.79 – 1.57	0.15 - 0.39	0.17 – 0.92	61.5 - 191.0	10.7 – 28.0	4.6 – 7.4	87.0 – 486.5

Table 5

Trichodesmium spp. densities (as trich. L^{-1}) literature review (Note: references considering colony abundance are not presented. Specific difference between *T. erythraeum* (§) and *T. thiebautii* (*) are reported when available. m: mean value; max: maximum value.

Region	Depth	Density	Author(s)
Sargasso and Caribbean seas	0 m (range) 15-200 m (range)	0-49* 0-294*	Carpenter and Price (1977)
Kuroshio (Japan)	0-100 m (max)	40 – 50000	Saino and Hattori (1980)
Great Barrier Reef lagoon	bloom (max)	50,000	Revelante and Gilmartin (1982)
Cleveland Bay (Australia)	inshore "blooms" (m) offshore "blooms" (m)	8515§ 3300§	Jones (1992)
HOT Station (Hawaii)	0-45 m (range)	11 – 84	Letelier and Karl (1996)
Southwest tropical Pacific	0 m - Fiji (max) 0 m New Caledonia (max)	10,000 1000	Dupouy et al. (2000)
Tanzania coast	0 m "bloom" (range) 0 - 20 m (range)	38,000 - 120,000§ 0 - 63,000§	Lugomela et al. (2002)
Gulf of Aqaba	0-90 m (range)	0.05 - 2	Post et al. (2002)
South China Sea	0 m (m, range)	77 (0 - 962)	Chen et al. (2003)
Magnetic island (Australia)	surface (range) 15 m (m)	9 - 102,000 13,000	Muslim and Jones (2003)
North and South Atlantic	7 m (m) (max)	300 2200	Tyrell et al. 2003
Dampier Archipelago (Australia)	0-20 m (m)	1800§	Negri et al. (2004)
North tropical Atlantic	"surface" (m) "surface", peak period (m)	222 – 292* 2250 (up to 10867)*	Carpenter et al. (2004)
Bay of Ouinné (New Caledonia)	0 - 45 m (m)	0.7 - 17§ ; 0.6 – 29*	Tenório et al. (2005)
Loyalty channel (New Caledonia)	0 - 60 m (range)	0 - 1011§ ; 4 – 2450*	Tenorio (2006)
Bay of Sainte Marie (New Caledonia)	0.5 m (range) 6 m (range)	0 - 240§ ; 0 – 34* 0 - 47§ ; 0 – 18*	Present study

Table 6

Literature review of *Trichodesmium* carbon-specific doubling times (d).

Reference	Conditions		Temperature (°C)	Doubling time (d)
Carpenter and Romans (1991)	natural populations, tropical -subtropical Atlantic		26.5 - 28	1.8 - 18
Carpenter and Capone (1992)	Review		>27	3 - 6
Carpenter et al. (1993)	natural populations	*T. erythraeum*	26.5 - 28	3.8
		T. thiebautii		3.0
Prufert-Bebout et al. (1993)	Cultures			3.0*
Mulholland and Capone (2000)	Cultures			2.2
	natural populations			3.8 - 198
Orcutt et al. (2001)	natural populations, Bermuda			2
Bell et al. (2005)	Cultures		25 ± 3	2.3 – 3.5
Mulholland and Bernhardt (2005)	continuous culture		28	3 - 10
Le Borgne et al. (2006)	natural populations, bloom, Bay of Ste Marie,		27 - 29	4.1 - 5.8
	idem, non-bloom conditions		27 - 29	13.9 - 40.8

* inferred from their 0.23 division d^{-1} growth rate

Table 7

Values of the trichome exponential net growth rate ($g_{0.5}$) and their corresponding doubling time ($d_{A, 0.5}$) satisfying equation 4, for z = 0.5 m. Percent contributions of net growth ($G_{0.5} / \Delta V_{0.5}$) and ascent from deeper levels ($A_{0.5} / \Delta V_{0.5}$) to observed biovolume increases at 0.5 m, $\Delta V_{0.5}$.

Period	$g_{0.5}$ (d^{-1})	$d_{A, 0.5}$ (d)	$A_{0.5}/ \Delta V_{0.5}$ (%)	$G_{0.5}/ \Delta V_{0.5}$ (%)
24-28 Nov 2003	0.18	3.86	87.3	12.5
3-5 Dec 2003	0.38	1.82	97.6	2.4
10-12 Dec 2003	0.12	5.57	87.3	12.6
13-15 Dec 2004	1.32	0.53	98.2	1.8
3-5 Jan 2005	0.11	6.45	87.0	13.0
25 Mar - 4 Apr 2005	0.17	4.14	98.9	1.1

Figure captions

Fig. 1. Sampling station locations at the entrance (Sta. SM) and off (Sta. O) the Bay of Ste Marie.

Fig. 2. *Trichodesmium* abundance as trichome numbers versus biovolume during the 2003 and 2004-2005 surveys. Slopes (b) of the linear regressions are : b= 44240 in 2003 and b= 45084 in 2004-2005, respectively.

Fig. 3. Temporal variations in *T. erythraeum* biovolume at Sta. SM four sampled depths, during Survey I (2003). Note : *T. thiebautii* densities, contributing for <1% of the total, have not been represented on the figure.

Fig. 4. Temporal variations in *T. erythraeum* and *T. thiebautii* biovolume at Sta SM two sampled depths, during Survey II (2004-2005). Less frequent observations were made at Sta. O, 0.5 m

Fig. 5. Relationships between trichome in colony (y) and free trichome (x) densities during the 2003 and 2004-2005 surveys.

Fig. 6. Temporal variations of environmental variables and *Trichodesmium* spp. densities at Sta. SM during Survey I. Temperature and salinity refer to the 1 m depth. Nitrate (NO_3), Soluble Reactive Phosphorus (SRP) and chlorophyll *a* (Chl *a*) concentrations are averaged through the 0-12 m water column, wind velocities, on 24 h. Significant rainfall (>10mm d^{-1}) are indicated by vertical arrows. *Trichodesmium* spp. abundances are integrated over 10m.

Fig.7. Same as Fig. 3, but for Survey II.

Fig. 8. *Trichodesmium* peak amplitude versus nitrate concentration at the beginning of the five blooms.

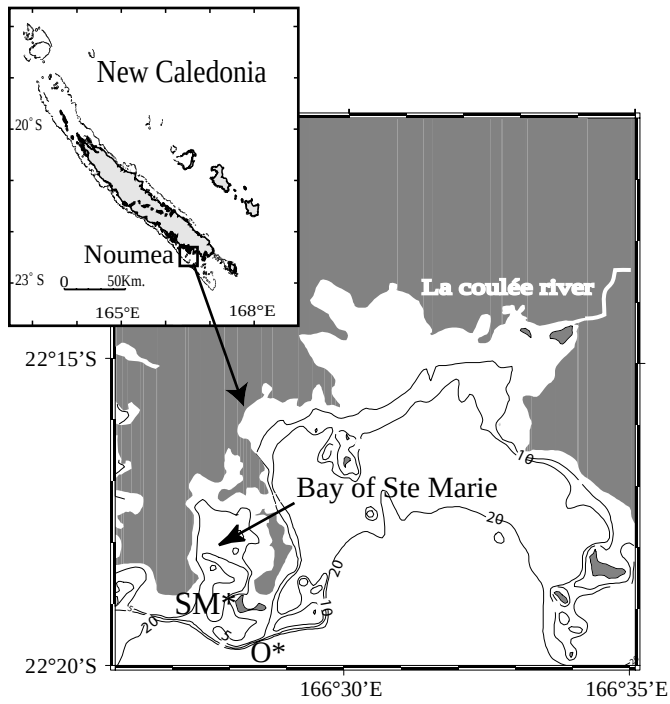
New Caledonia
20° S
Noumea
23° S
0 50Km.
165°E
168°E
La coulée river
22°15'S
Bay of Ste Marie
10
20
20
20
10
SM*
O*
22°20'S
166°30'E
166°35'E

Fig. 1

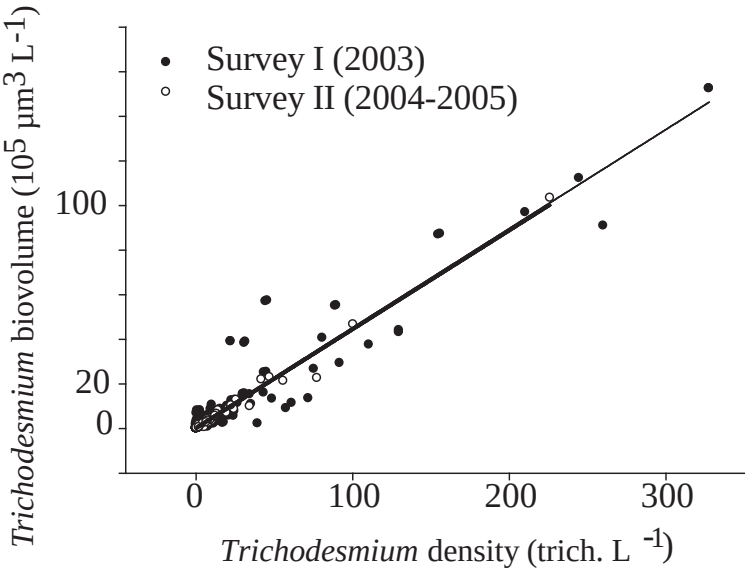
Survey I (2003)
Survey II (2004-2005)
Trichodesmium biovolume (10^5 µm^3 L^{-1})
100
20
0
0
100
200
300
Trichodesmium density (trich. L^{-1})

Fig. 2

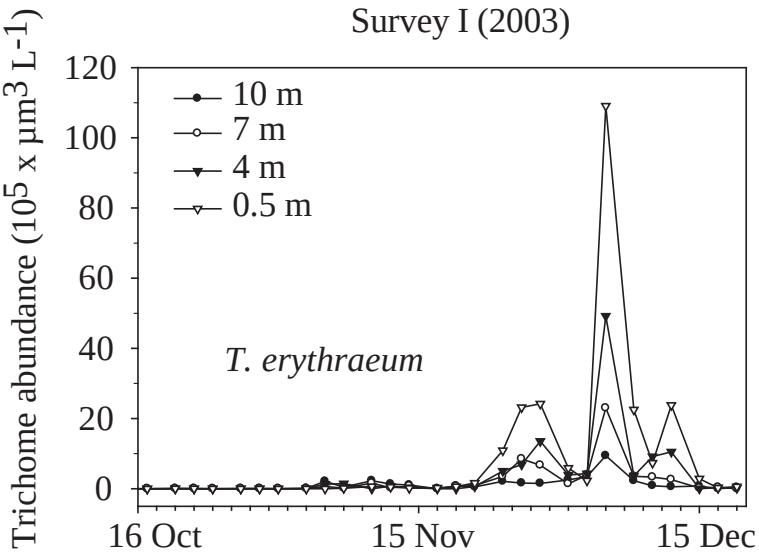
Survey I (2003)
Trichome abundance (10^5 x µm^3 L^-1)
120
100
80
60
40
20
0
10 m
7 m
4 m
0.5 m
T. erythraeum
16 Oct
15 Nov
15 Dec

Fig. 3

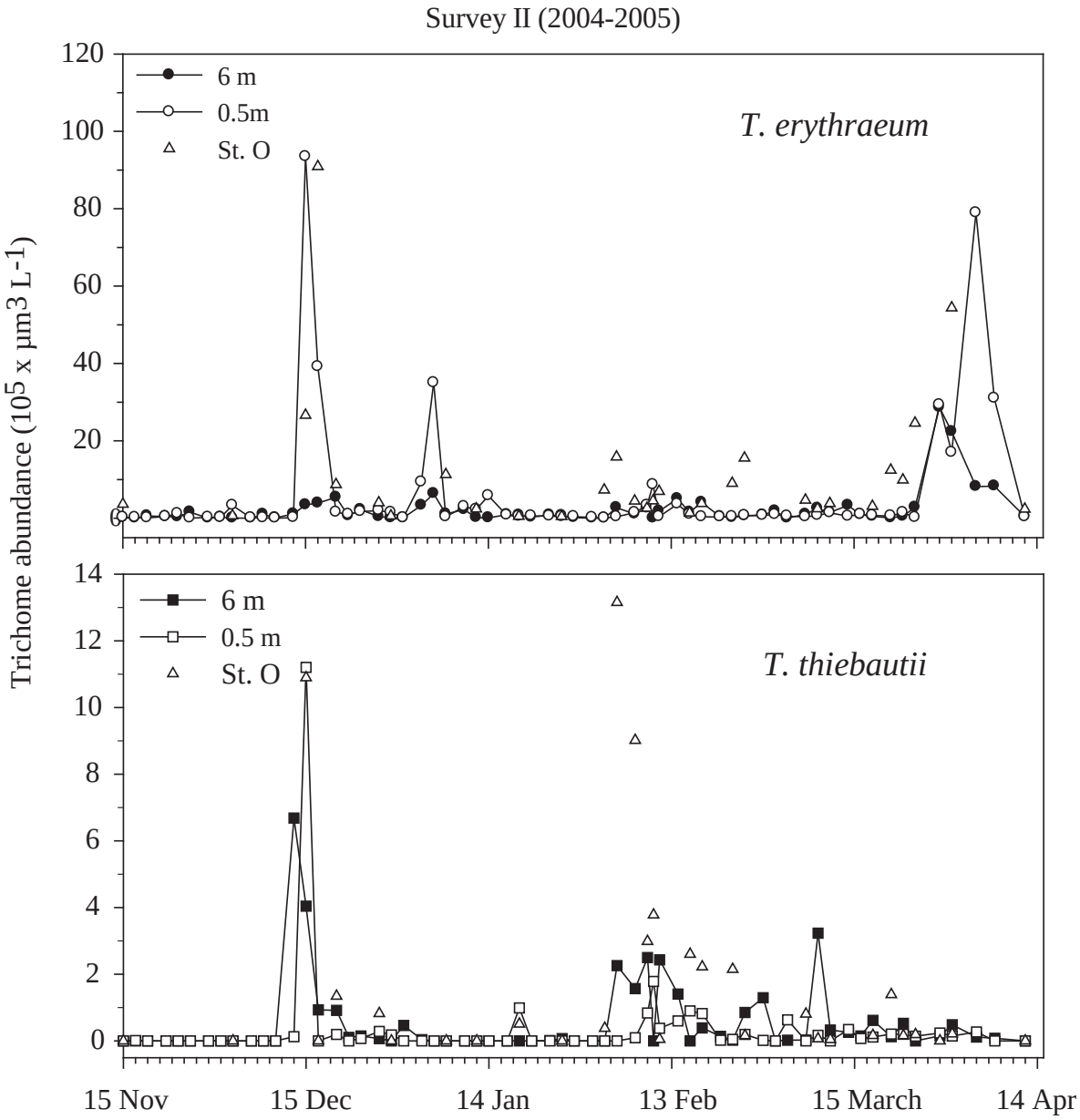
Survey II (2004-2005)
Trichome abundance (10^5 x µm^3 L^-1)
6 m
0.5m
St. O
T. erythraeum
6 m
0.5 m
St. O
T. thiebautii
120
100
80
60
40
20
14
12
10
8
6
4
2
0
15 Nov
15 Dec
14 Jan
13 Feb
15 March
14 Apr

Fig. 4

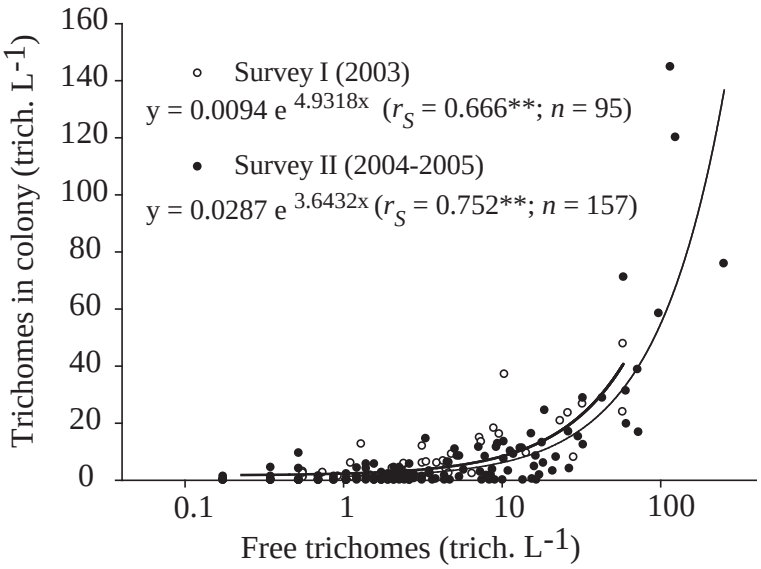
Trichomes in colony (trich. L-1)
160
140
120
100
80
60
40
20
0
Survey I (2003)
y = 0.0094 e 4.9318x (rS = 0.666**; n = 95)
Survey II (2004-2005)
y = 0.0287 e 3.6432x (rS = 0.752**; n = 157)
0.1
1
10
100
Free trichomes (trich. L-1)

Fig. 5

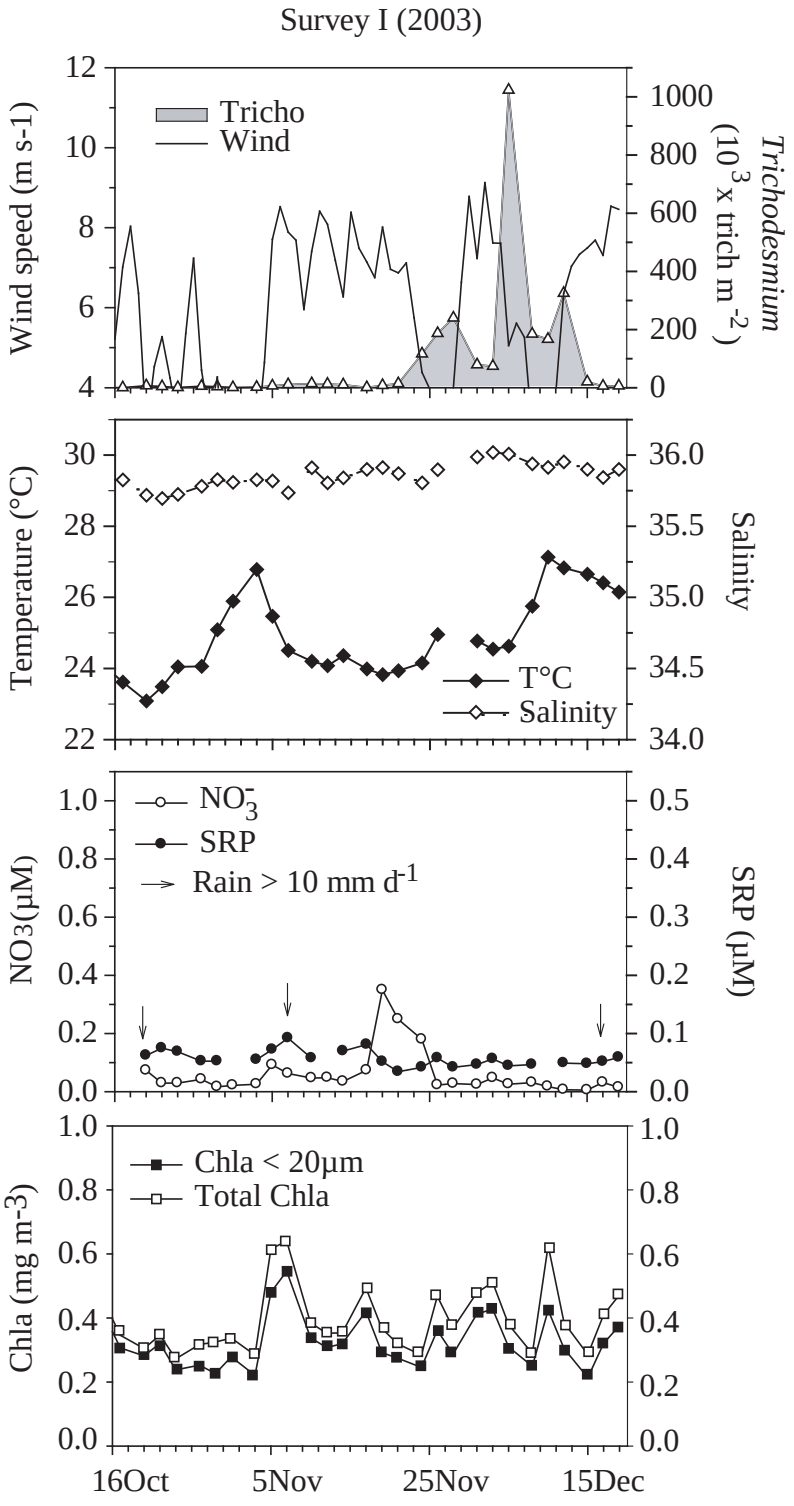
Survey I (2003)
Wind speed (m s-1)
Tricho
Wind
Trichodesmium
(10 3 x trich m -2)
Temperature (°C)
T°C
Salinity
NO3(µM)
NO3-
SRP
Rain > 10 mm d-1
SRP (µM)
Chla (mg m-3)
Chla < 20µm
Total Chla
16Oct
5Nov
25Nov
15Dec

Fig.6

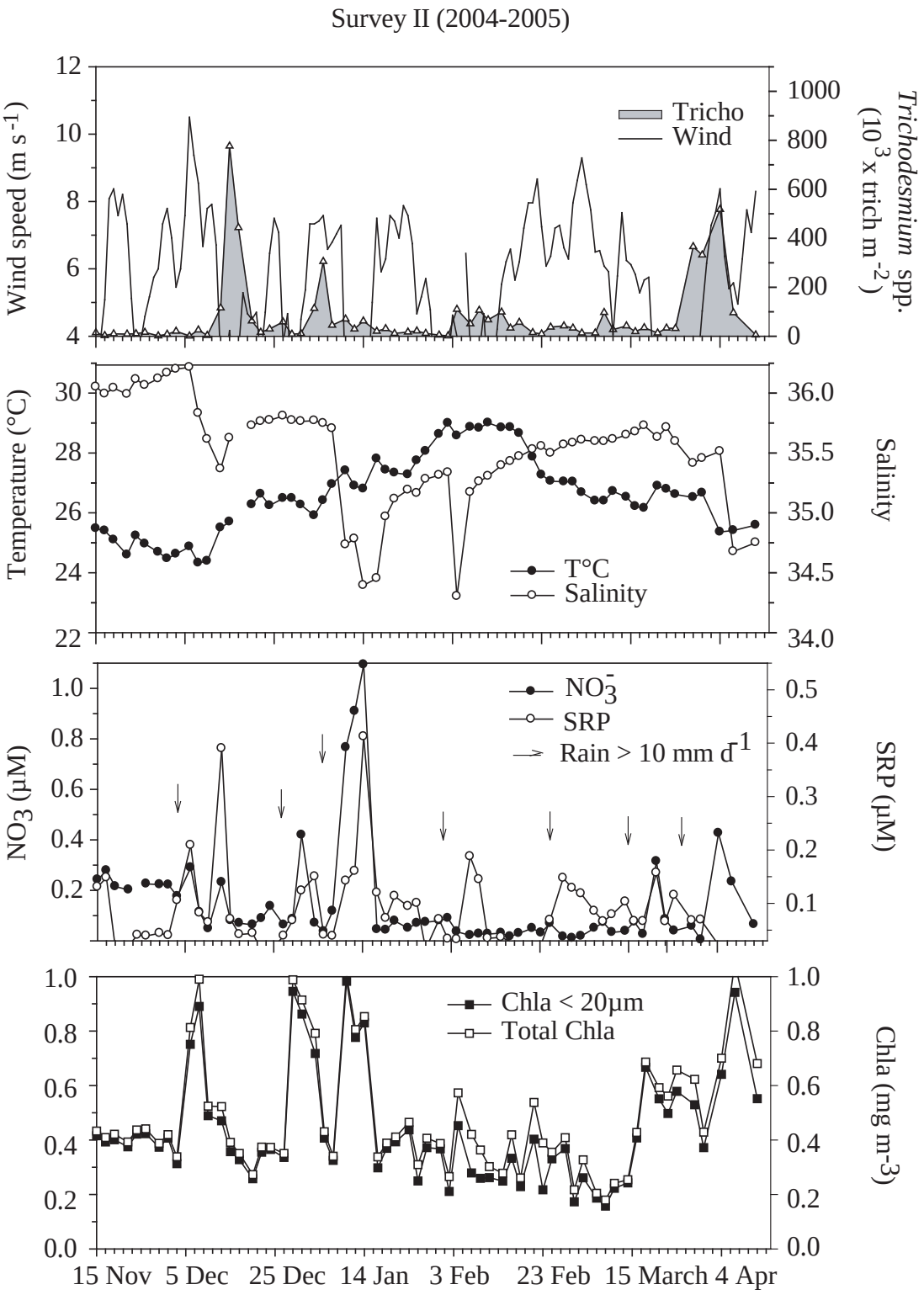
Survey II (2004-2005)
Wind speed (m s-1)
Tricho
Wind
Trichodesmium spp.
(10 3 x trich m -2)
Temperature (°C)
T°C
Salinity
Salinity
NO3 (µM)
NO3-
SRP
Rain > 10 mm d-1
SRP (µM)
Chla < 20µm
Total Chla
Chla (mg m-3)
15 Nov
5 Dec
25 Dec
14 Jan
3 Feb
23 Feb
15 March
4 Apr

Fig. 7

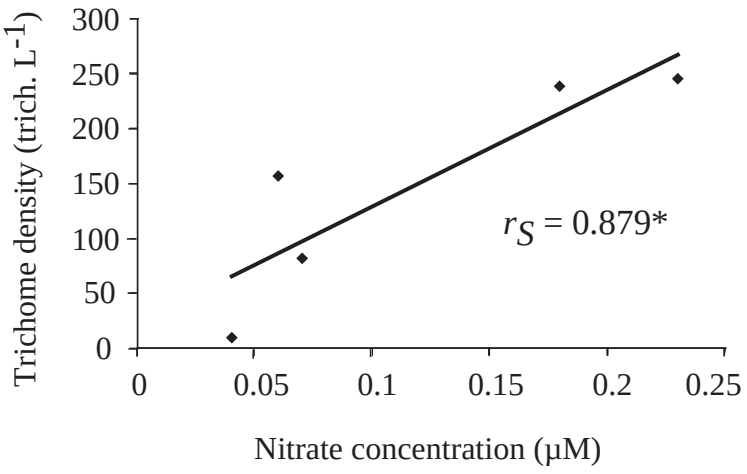
Trichome density (trich. L-1)
300
250
200
150
100
50
0
0
0.05
0.1
0.15
0.2
0.25
Nitrate concentration (µM)
rS = 0.879*

Fig. 8