Malaysian Journal of Science **25** (2): 67 – 79 (2006)

Photosynthetic Light Responses of Wild and Cultured *Halophila ovalis*

Mohammad Rozaimi Jamaludin*, Japar Sidik Bujang, Misri Kusnan,
Hishammuddin Omar
Department of Biology, Faculty of Science, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor, Malaysia
*entropicity@yahoo.com

ABSTRACT A laboratory based experiment was performed on leaves of the seagrass *Halophila ovalis* to elucidate its photosynthesis versus irradiance (P-I) relationships. Plant samples were sourced from a low water level seabed off the Teluk Kemang coast (2 ° 30'N, 101 ° 45'E) in Negeri Sembilan, Malaysia. Plants growing naturally from this area as well as those transplanted into indoor culture tanks were studied. The oxygen evolution responses of the leaves based on parameters of leaf fresh weight (FW), leaf surface area (Area) and leaf chlorophyll content (Chl) towards varying degrees of illumination were recorded. A comparison between leaves from cultures with those from the wild showed that the curve plotted for its respective values was lowered for the former based on FW and Area but comparatively lowered based on Chl. The light compensation (I_c) value did not vary much between leaves from the wild and leaves from the cultures (8-13 μmol m^{-2} s^{-1}) while light saturation point (I_k) was in the range of 268 - 275 μmol m^{-2} s^{-1} for leaves from the wild and increased to 290 - 293 μmol m^{-2} s^{-1} for leaves from cultures. Dark respiration values did not differ between wild and cultured leaves based on the measurements calculated from the parameters stated (FW, Area and Chl). Calculations based on FW and Area showed a higher light saturation (P_{max}) photosynthetic rate for cultured leaves but comparatively lowered based on Chl. P_{max} values between leaves from culture to that from the wild based on the parameters of FW and Area were significantly different ($p<0.05$) but not based on Chl while the trends of curves fitted between wild and cultured leaves were significantly different based on Area only ($p<0.05$). The results collated in this study serve to add to the present knowledge of biological traits of *H. ovalis* from Malaysia to further understand its importance in local waters.

ABSTRAK Satu ujikaji makmal telah dijalankan ke atas daun *Halophila ovalis* untuk menjelaskan perkaitan antara kadar fotosintesisnya dengan faktor keamatan cahaya. Tumbuhan kajian diambil dari habitat asalnya di perairan pantai Teluk Kemang (2° 30'N, 101° 45'E), Negeri Sembilan, Malaysia. Daun daripada tumbuhan yang dikultur dan tumbuhan liar telah digunakan untuk kajian tersebut. Tindak balas evolusi oksigen terhadap peringkat keamatan cahaya yang berbeza bagi daun telah direkodkan adalah berdasarkan tiga parameter – berat segar daun (FW), luas permukaan daun (Area) dan kandungan klorofil daun (Chl). Perbandingan nilai antara daun tumbuhan liar dengan tumbuhan yang dikultur dibuat berdasarkan graf yang diplotkan. Graf menunjukkan keluk nilai yang diplotkan berdasarkan FW dan Area adalah lebih rendah berbanding keluk yang diplotkan berdasarkan Chl. Tiada perbezaan ketara antara nilai kompensasi cahaya bagi daun tumbuhan liar dan daun tumbuhan yang dikultur (8-13 μmol m^{-2} s^{-1}). Manakala nilai ketepuannya ialah 268 - 275 μmol m^{-2} s^{-1} bagi daun tumbuhan liar dan meningkat sehingga 290 - 293 μmol m^{-2} s^{-1} bagi daun tumbuhan yang dikultur. Tiada perbezaan ketara bagi kadar respirasi gelap antara daun liar dengan daun kultur mengikut pengukuran kadar respirasi yang dibuat berdasarkan parameter yang dinyatakan (FW, Area dan Chl). Pengiraan berdasarkan FW dan Area menunjukkan nilai kadar ketepuan cahaya fotosintetik (P_{max}) yang tinggi bagi daun tumbuhan yang dikultur tetapi lebih rendah jika dibandingkan nilai pengiraan berdasarkan Chl. Terdapat perbezaan ketara bagi P_{max} di antara daun yang dikultur dan daun liar berdasarkan parameter FW dan Area ($p<0.05$) tetapi tiada perbezaaan ketara bagi P_{max} yang berdasarkan Chl. Bagi plot yang dihasilkan, lengkung di antara daun kultur dan daun liar hanya mempunyai perbezaan ketara berdasarkan Area sahaja ($p<0.05$). Keputusan yang dicerap daripada ujikaji ini penting untuk memahami ciri-ciri *H. ovalis* dan kepentingan rumput laut tersebut di perairan Malaysia.

(Seagrass, *Halophila ovalis*, Photosynthesis-irradiance curves, Shading)

INTRODUCTION

Seagrasses grow in a variable environment that may affect their proliferation and reproduction. Environmental conditions fluctuate primarily according to the availability of substrate, nutrients and light. Responses are essential to counter any changes in its relatively unpredictable marine habitat as shown by plants in artificial culture experiments. These responses are very much inducible.

Seagrasses such as *Cymodocea serrulata* [1], *Cymodocea nodosa* [2], *Zostera capricorni* [3], *Syringodium isoetifolium*, *Halodule uninervis* and *Halophila spinulosa* [4] have demonstated various changes in morphology from artificial manipulations of various environmental parameters. *In situ* nutrient manipulations on *Halodule wrightii* [5] have also been shown to elicit morphological differences from plants growing in the same jurisdictions. Differing manipulations in light intensity levels investigated upon natural growths of *Posidonia oceanica* [6], *Thalassia testudinum* [7] and *Zostera marina* [8] demonstrated that reduced light was tolerable until its thresholds to photo-acclimatize were crossed.

Light is one of the deterministic factors of seagrass proliferation and photosynthetic capacities would also vary significantly within the same species. Basic plant physiology explains the fundamental role of photosynthesis being the mechanism by which carbohydrates are converted from atmospheric carbon dioxide and water in the presence of photosynthetic pigments and light as an energy source. Oxygen is produced as a by-product, which is approximately proportional to the whole photosynthetic activity. It is thus a conventional approach to estimate photosynthetic productivity of any plant by the amount of oxygen evolved. In seagrasses, a considerable amount of the oxygen produced is transported to the tissues below-ground (underground rhizomes and roots). This physiological process is of minor importance to terrestrial plants. The sediment of seagrass beds is usually anoxic and the oxygen supply of the roots and rhizomes are dependent on an oxygen flux from the shoots to the organs below-ground [9]. Hence, the oxygen available to the roots and rhizomes is largely derived from photosynthesis. This is conveyed by gaseous diffusion through internal gas spaces – lacunae - which extends from the leaf to the root apices [10].

Various methods have been employed to measure the oxygen evolved by seagrasses based on its high diffusibility in aqueous media. Different plant parts have been experimented upon. Some studies were based on portions of leaf blades [11-13], while others used single plant units - either by placing sealed transparent chambers around the photosynthetic leaves [14, 15] or by measuring lacunal gas discharge from the base of the erect shoot [16]. Methods approximating photosynthetic rates from whole plant canopies upon naturally occurring seagrass beds were also used [17, 18].

All the methods employed were aimed to illustrate a plant's productivity values - usually in terms of photosynthesis versus irradiance (P-I) relationships of the plant to light exposure, whether of terrestrial or aquatic origin. The relationship between plants' photosynthesis and their direct environment essentially endeavours to elucidate the plant's compensation point, saturation point and photoinhibition point, although, the latter value is seldom investigated for seagrasses [9].

This study is thus intended to determine the range of these photosynthetic values in addition to the maximum photosynthetic rate, dark respiration rate and photosynthetic efficiency of *H. ovalis* from Malaysia - a common seagrass found locally. This forms one of the most important productivity values which could explain trends in its basic biological demands for growth. This aim is further expanded to compare these values between plants growing naturally in the wild with those cultured in light-controlled tanks. As a continuation of previous studies on the effects of light attenuation on *H. ovalis* [19, 20], the results collated will be able to provide a better understanding of the characteristics of this seagrass in local environments in Malaysia.

MATERIALS AND METHODS

Plant material

Two types of plant samples were used for the study - those growing naturally in the wild and those originally collected from the wild and transplated into culture tanks. Plants were collected from a seagrass bed off Teluk Kemang coast (2 ° 30'N, 101 ° 45'E) during low tide. For cultures, single sprigs of *Halophila ovalis* from the same area were transplanted in baskets in October of 2004 and placed in a 200 l capacity tank. Aeration was provided to produce water circulation. Seawater of salinity 30 psu was obtained from the nearby sea. Natural sunlight provided for the plants was reduced by a commercially available shade cloth. The shading resulted in light intensity attenuated to about 150 - 250 µmol m^{-2} s^{-1} providing approximately about 10 - 15% of total light intensity outdoors. The ambient temperature of the culture tank ranged from 26.5°C – 29.0°C. Prior to the study, these culture conditions had been maintained for twelve months since October 2004.

Experimental mechanism

Leaves for the study were obtained and studied within a few hours before performing the experiment for plants in cultures while leaves from wild plants were experimented upon within twelve hours upon obtaining them from its habitat. The time interval between removal from the culture tank or original habitat was kept to a minimal to reduce any physiological changes that might have occurred within the leaf samples. Leaves free from epiphytes were selected for the experiment and placed in a 5 ml by volume incubation chamber fitted to a Clark's type oxygen incubation electrode (Rank Brothers). Oxygen evolved by photosynthesis from the leaves incubated will diffuse through a thin Teflon membrane below the incubation chamber and is reduced at a platinum surface immediately in contact with the membrane. The oxygen produced is proportional to the light intensity provided. The light intensity can be varied by moving light source at different distances with respect to the incubation chamber. Temperature of the incubation chamber was monitored and kept constant by flowing tap water. The ambient water temperature ranged from 26.5°C – 29.0°C but was controlled at one temperature during the experiment in accordance to the environment at the time the experiment was performed. Data output was recorded by a potentiometric recorder (Zipp and Konnen).

For wild *H. ovalis*, two leaves were placed in the incubation chamber to produce detectable responses. For leaves in culture, a single leaf was sufficient for measurable responses since cultured leaves had grown much larger in surface area than those from the wild [19]. Five replicates were performed from both types of leaf specimens. The light source was a strong halogen bulb illuminated in increasing irradiance grades from 30 µmol m^{-2} s^{-1} to 400 µmol m^{-2} s^{-1}. In its natural environment, *H. ovalis* is seldom exposed to intensities greater than this range unless during extreme low tides (mid-day light intensities is in the range of 100 - 400 µmol m^{-2} s^{-1} at submerged depths of 4 and 2 m respectively). Light intensities beyond this range were not used as a precaution to prevent any internal photodamage to the leaf. Futhermore, it had been shown that exposures to high light intensities will result in notable changes in the pH of the seawater surrounding *H. ovalis* leaves [21]. The photosynthetic rate was then calculated based on leaf fresh weight, leaf surface area and chlorophyll content.

Leaf parameters

For fresh weight, each of the leaves were dried by blotting and weighed. The leaf surface area was estimated by tracing its outline on a graph paper. For calculating chlorophyll content, leaf blades were minced and soaked in 5 ml of 80% acetone. Absorbances of the extracts were then measured in a spectrophotometer at 725, 664 and 647 nm wavelengths [see 22]. Chlorophyll *a* and *b* concentrations were calculated from these absorbances using the appropriate equations [23].

Graphical and statistical analyses

The results of oxygen evolution with respect to its corresponding light irradiance level were then plotted on graphs. These graphs plotted would display a rectangular hyperbola shape of a photosynthetic – irradiance (P-I) curve. The conspicuous points to note are as follows. Where the curve crosses the light intensity axis, this point is the compensation point - denoted as the irradiance where gross photosynthesis equals respiration and net photosynthesis is zero (I_c). Where the curve extends to cross the photosynthetic rate axis, the dark respiration point (R_{dark}) - designating the net consumption of oxygen as a consequence of respiration in the dark which is generally assumed to remain constant in the light. The symbol α represents the photosynthetic efficiency, i.e. the ratio between photosynthesis rate and irradiance which is approximately a linear function of irradiance. The steeper the slope, the more efficient that plant is to increasing levels of low irradiance. The plateau formed at a higher irradiance level signifies the light saturated rate (P_{max}) - the maximal photosynthetic capacity of the leaf. The saturation point/saturation irradiance (I_k) is given as the intercept between α and the plateau. Theoretically, if the curve is extended beyond the plateau, there will be a downward progression of the slope; the intercept between this slope and the plateau representing the photoinhibition point (P_I). However, this is rarely investigated for seagrasses [9].

From the values obtained, the graphical trends between the leaves from culture and those from the wild are differentiated. The photosynthetic values of the leaves will then be inferred. Figure 1 demonstrates the photosynthetic points of I_c, I_k, P_{max}, R_{dark} and α being described.

For statistical analyses (NCSS software package), a Student's T-test was used to assess the significance of differences in photosynthetic values between cultured and wild *Halophila ovalis* leaves. Graphs of the P-I relationship were plotted and the trends were assessed using regression. A Monte Carlo method was used to test the hypothesis that the curves fitted between wild and cultured leaves were significantly different.

RESULTS

Experimental photosynthetic light responses

The curves fitted showed that the P-I responses varied between leaves from the wild with those from cultures for all three parameters of leaf fresh weight (FW), leaf surface area (Area) and leaf chlorophyll content (Chl). Values plotted against leaf fresh weight and leaf surface area (Figures 2a and b respectively) showed that there was an apparent elevation of the curve for the photosynthetic rate of cultured leaves compared to wild leaves (Figure 2c). Comparatively, the curve based on leaf chlorophyll content was slightly higher for photosynthetic rates of wild leaves than cultured leaves. The plots were fitted according to Michalis-Menten kinetics trends using non-linear regression ($r^2 > 0.979$ for all curves) based on the equation:

$$P = [(P_{max})(I)]/(K_m + I)] - R_{dark}$$

where K_m is the arbitrary half-saturation constant and I is the irradiance in μmol m^{-2} s^{-1} [following 24].

Using analyses of Student's T-test, significant differences between photosynthetic rates of wild leaves and cultured leaves were observed at the light intensity of 400 μmol m^{-2} s^{-1} only for parameters based on Area and FW. Hence, P_{max} is statistically different based on Area and FW ($p<0.05$). There were no significant differences between the two types of leaves based on Chl. The Monte Carlo test showed that the curves between the two types of leaves were significantly different for oxygen evolution rate based by Area but not by FW or Chl.

The values for I_c and R_{dark} are very similar between wild leaves and cultured leaves based on all three parameters of FW, Area and Chl. Comparatively, I_k values increased by about 7.33 % (an increase of about 20 μmol m^{-2} s^{-1}) for cultured leaves. P_{max} and photosynthetic efficiency, α, values also differed according to the base parameter. The largest difference for P_{max} was observed from the photosynthetic rate based on Area (cultured leaves increased by 23.65%) while the largest difference for α was based on Chl (cultured leaves decreased by 11.38%). Table 1 summarises the photosynthetic rates as inferred from their respective curves, while Table 2 compares some photosynthetic irradiance values (I_c and I_k) of *Halophila* species.

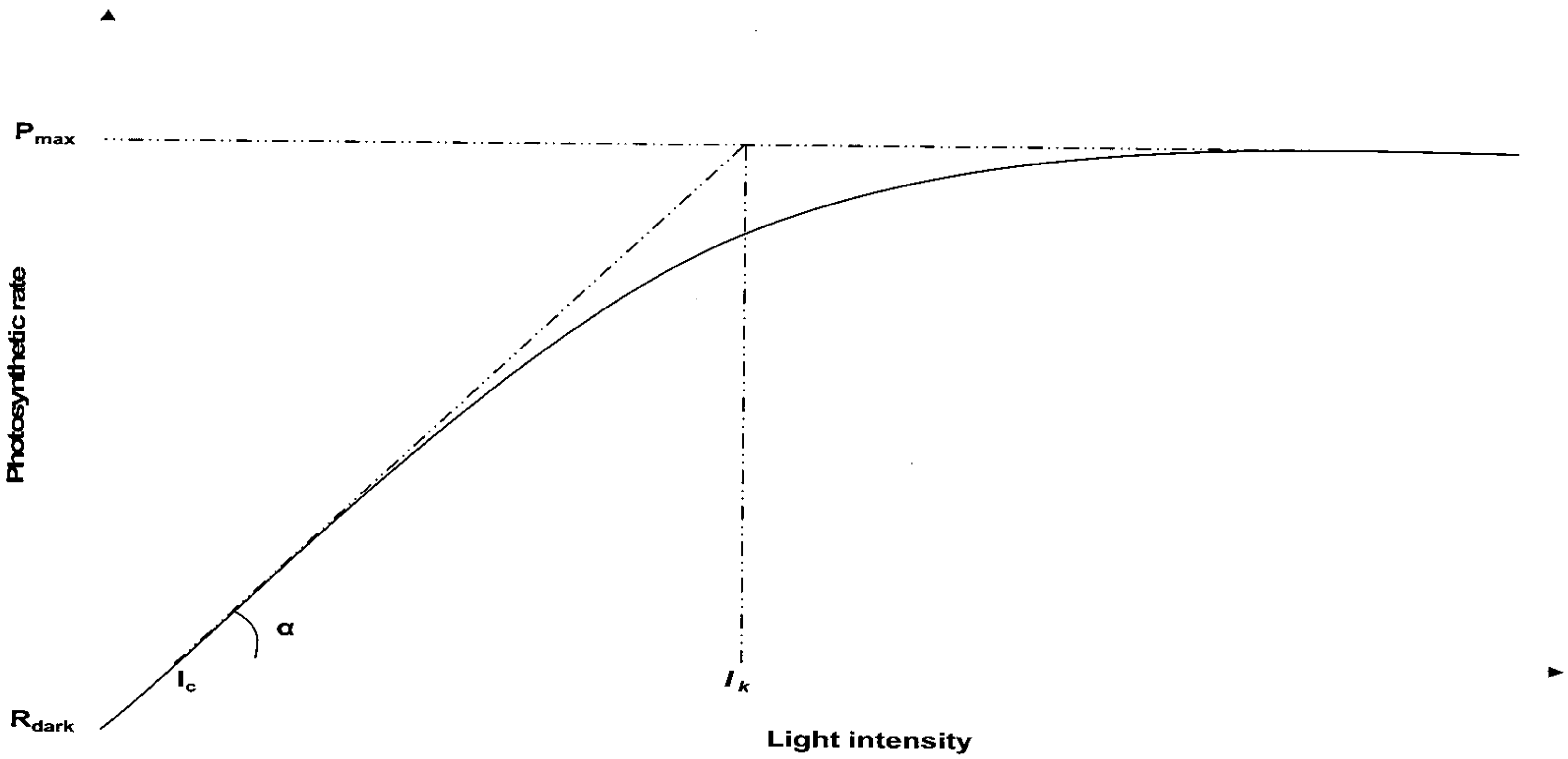

Figure 1. Theoretical progression of a Photosynthesis-Irradiance (P-I) curve. The points to note are: P_{max} - the maximal photosynthetic rate; I_c - irradiance where gross photosynthesis equals respiration and net photosynthesis is zero; I_k - saturating irradiance R_{dark} - net consumption of oxygen from respiration in the dark; α - the photosynthetic efficiency, calculated as the ratio between photosynthesis rate and irradiance.

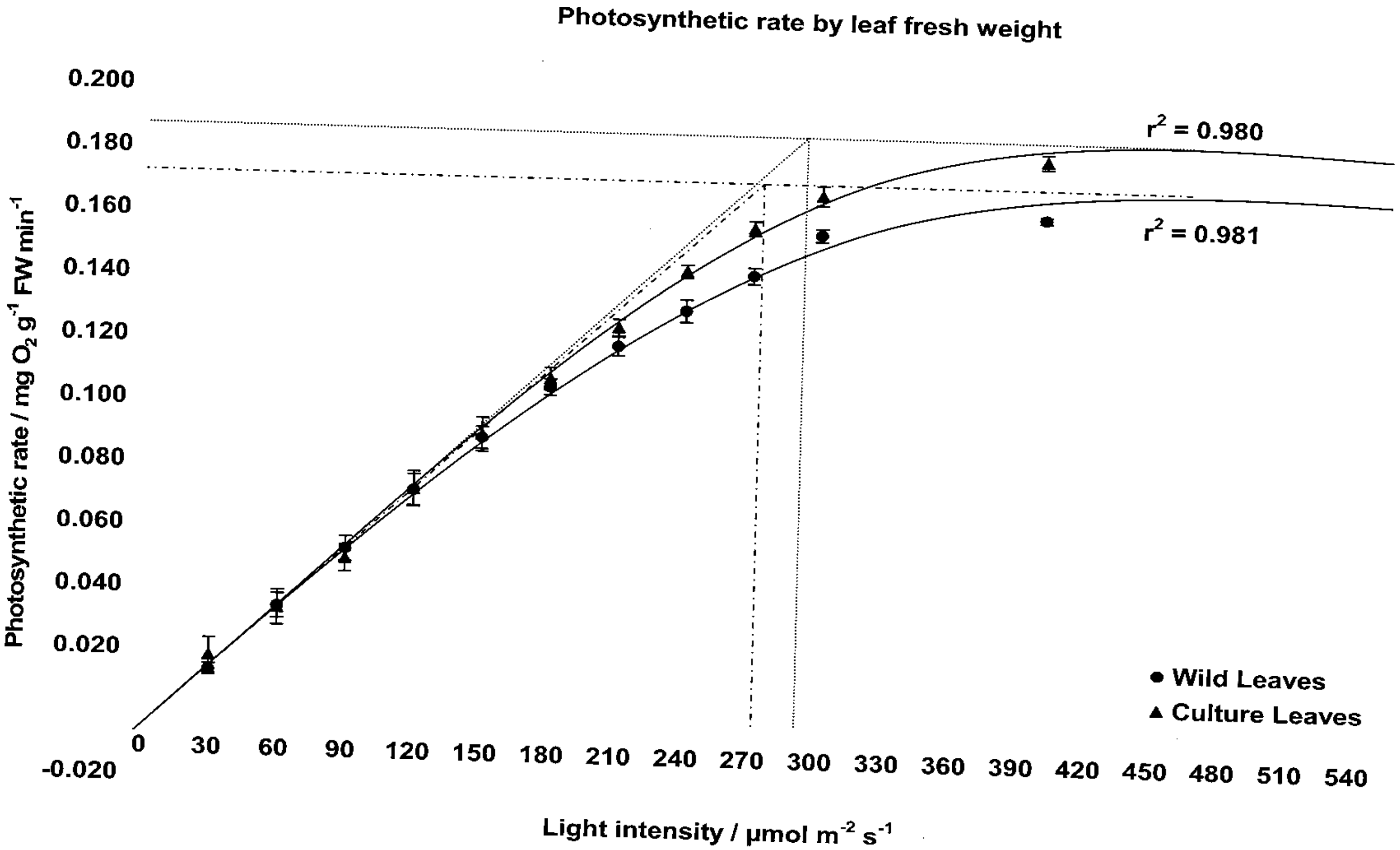

Figure 2a. Photosynthetic rates based on leaf fresh weight.

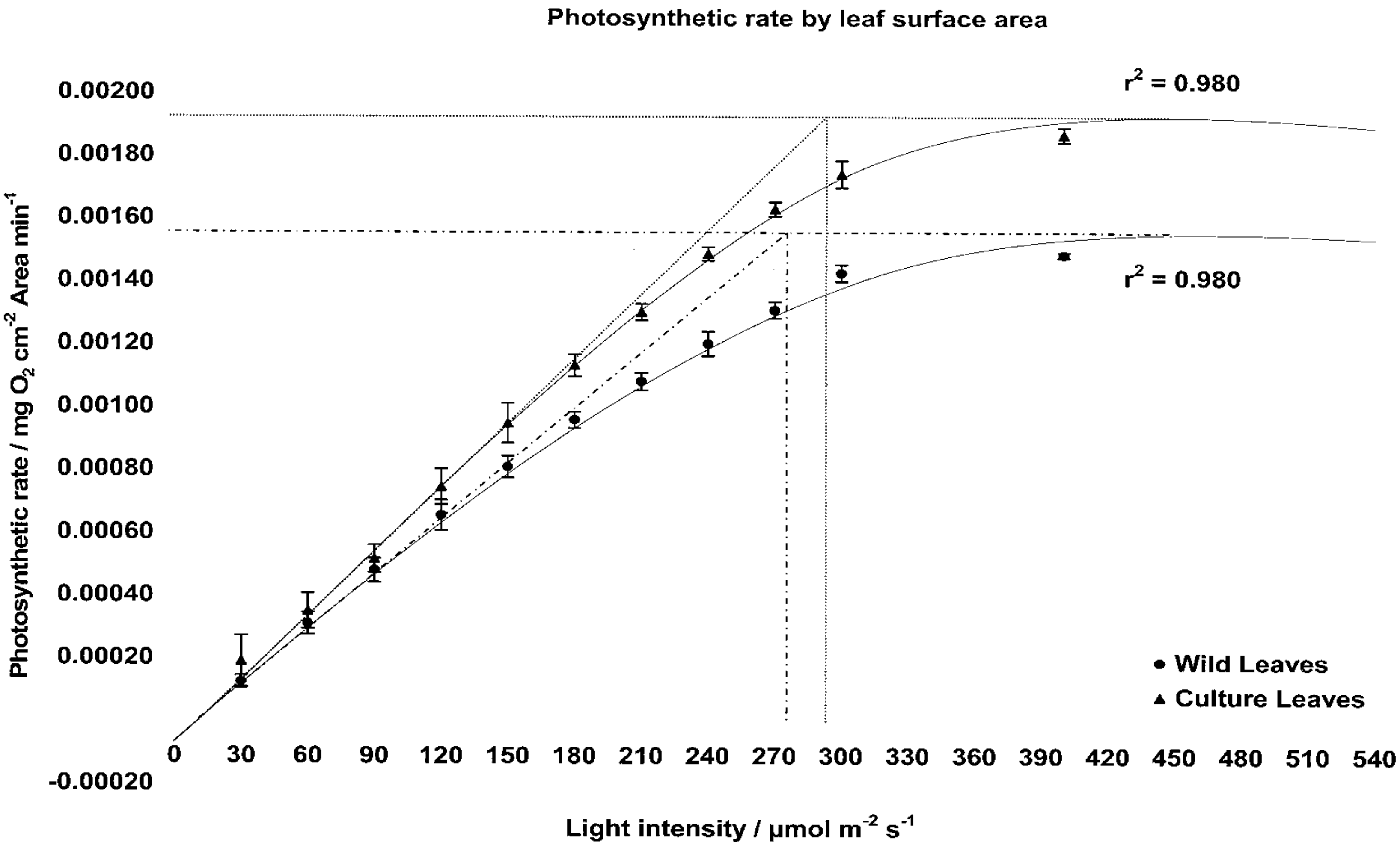

Figure 2b. Photosynthetic rates based on leaf surface area.

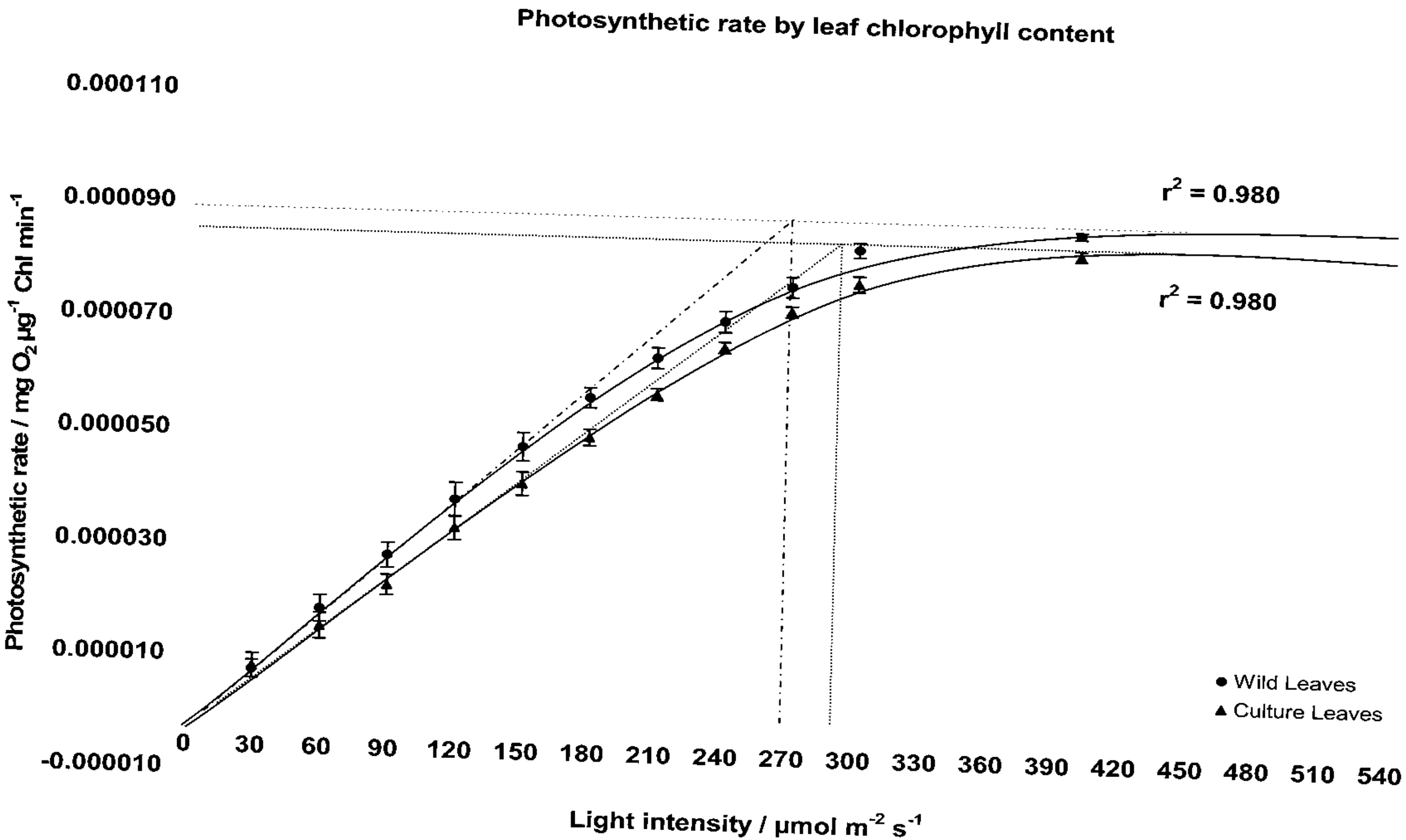

Figure 2c. Photosynthetic rates based on leaf chlorophyll content.

Table 1. Summary of the photosynthetic rates (R_{dark}, I_c, I_k and P_{max} values) inferred from their respective curves. I_c and I_k are measured in μmol m^{-2} s^{-1}. R_{dark} and P_{max} are each measured by its respective base parameters- leaf fresh weight (mg O_2 g^{-1} FW min^{-1}); leaf surface area (mg O_2 cm^{-2} Area min^{-1}); and leaf chlorophyll content (mg O_2 μg^{-1} Chl min^{-1}). α, calculated as P_{max} / I_k, is arbitrarily taken as photosynthetic rate from its respective base parameter per light intensity. Percentage differences for P_{max} and α is the elevation or reduction of oxygen evolution rates of cultured leaves calculated with respect to wild leaves. Asterisk (*) denotes significant differences between leaves from the wild and leaves from cultures (student's T-test, $p<0.05$). $N = 5$.

PARAMETER	LEAF SOURCE	R_{dark}	I_c	I_k	P_{max}	P_{max} % DIFFERENCE	A	A % DIFFERENCE
By leaf fresh weight	Wild	0.00057	8	273	0.1725*	+ 8.75%	0.0006319	+ 1.33%
	Culture	0.00057	8	293	0.1876*		0.0006403	
By leaf surface area	Wild	0. 000065	10	275	0.001556*	+ 23.65%	0.00000566	+ 16.10%
	Culture	0.000065	10	293	0.001924*		0.00000657	
By leaf chlorophyll content	Wild	0.0000030	9	268	0.0000896	- 4.35%	0.000000334	- 11.38%
	Culture	0.0000037	13	290	0.0000857		0.000000296	

Table 2. Photosynthetic irradiance values (I_c and I_k) and its corresponding plant part used from selected *Halophila* by exposure to graded light regimes.

SPECIES	I_c	I_k	HABITAT DEPTH IN M	PLANT PART USED	LOCALE	SOURCE
Halophila ovalis	8 -13	268-293	0.5-1.0	leaf blade	Port Dickson, Malaysia	This study
Halophila ovalis	33	400	15	plant canopy	South Sulawesi, Indonesia	[18]
Halophila ovalis	40	200	2	whole shoots	Canning Estuary, Australia	[25]
Halophila stipulacea	20-40	300	10	leaf blade	Gulf of Eilat, Israel	[11]
Halophila decipiens	29	100-200 (inferred)	20	leaf blade	Florida, U. S. A.	[26]
Halophila johnsonii	50	300-400 (inferred)	0.3	leaf blade	Florida, U. S. A.	[26]
Halophila engelmannii	10 -60	432-504	1.0-2.5	compound leaf	Florida, U. S. A.	[27]

DISCUSSIONS

It has to be noted that the method employed to determine the rate of photosynthesis for *Halophila ovalis* was wholly laboratory based and that only the upper-ground leaves of the plant were used for the measurements. Field-derived photosynthetic parameters may display significantly different values from those determined from the laboratory [28, 29]. There may be several reasons [see 9 for further elaborations] for such discrepancies but chiefly, laboratory incubation temperatures and pressures are factors that have vital and immediate impact on photosynthetic parameters [11, 30, 31], which are usually not identical to *in situ* conditions. More importantly, experiments on individual leaves only reflect biological demands for that plant portion only. The respiratory requirements of the below-ground tissues will inevitably raise the light compensation point for photosynthesis in entire plants above that of the leaves only. For instance, the light compensation point of entire *Halodule wrightii* plants appeared to be 111 μmol m^{-2} s^{-1}, much higher than the 22 μmol m^{-2} s^{-1} determined as the light compensation for isolated leaf segments [28]. To this effect, Table 2 summarises some data on the photosynthetic irradiance values of several species in the genus *Halophila*. The compensation and saturation values of *H. ovalis* from Indonesia and Australia [18, 25 respectively] studied at a whole-plant level had been determined previously. There is only a slight difference in I_c values of *H. ovalis* from the two locales but quite large a difference in saturation values. Presumably, the main variability would be the depth factor (hence in terms of light attenuation). The other main abiotic factor could be due to the effects of pressure on photosynthesis, but it had been investigated that pressure had no significant effect on *Halophila* seagrasses [11]. If results from these studies are inferred with values from Australia and Indonesia, assuming that actual whole-plant I_c value from Teluk Kemang is approximately about that to these values, the supposed difference would be around 20 - 32 μmol m^{-2} s^{-1}- an elevation of almost 3 - 4 times to that of the 8 - 13 μmol m^{-2} s^{-1} determined from this study. Thus, it could be deduced that this increase in light intensity is needed to provide the additional energy needed to maintain the non-photosynthetic portions of the plant. Consequently, the energetic cost of sustaining these portions is relatively quite demanding on energy production. On the other hand, there is relatively a big difference when comparing the I_k values between the three study sites from Malaysia, Australia and Indonesia. Although further confirmations need to be made, it is likely that the I_k value recorded from this study is higher to that on the scale of a whole plant budget. It would be so since the value of 268 - 293 μmol m^{-2} s^{-1} produced the maximal photosynthetic capacity of the leaf organ of the plant only. It can also be said that this is the optimal photosynthetic capacity of the plant since no oxygen produced is transported for usage by the non-photosynthetic below-ground organs. After deductions from the utilisation of the oxygen produced used by the non-photosynthetic organs, this I_k value of a whole-plant budget would be projected to be of a lower value. If this was the case, the different locales have had a pronounced effect on I_k values rather than I_c. Environmental parameters of nutrients or sediment apart from light could be the causal factors for these differences.

The graphical patterns of the photosynthetic capacities of *H. ovalis* tend to vary as a result of transplanting into cultures; the prima facie would be due to the shading imposed. P–I investigations of this nature where the plant was maintained under different levels of light environment for a particular time period is more commonly done on algae [32, 33]. The trend is similar - curves of cultures preconditioned to low light tended to be elevated in terms of weight but lowered in terms of chlorophyll content. It is less common for this type of preconditioning treatment approach being applied to seagrasses. Some studies on *Zostera marina* [34] and *Posidonia oceanica* [35] were based on habitat depth difference or seasonality, but as mentioned, other environmental conditions must also be taken into due consideration. Nevertheless, the findings are similar as well.

Studies investigating photosynthetic values of seagrasses commonly use leaf weight - either fresh or dry, leaf surface area and chlorophyll content as the basal parameter. Additionally, parameters such as plant growth [36] or carbon production - measured directly or converted from amount of oxygen produced [34] were also employed in place of oxygen evolution. The choice of parameter to be used basically depends on the requirements on the overall physiological aspects being investigated. In the case of the effects of light reduction solely, the most appropriate base reference would be by chlorophyll content.

Chlorophyll content which reside in the chloroplast cells of the plant's photosynthetic portions is directly related to the amount of light it receives. Essentially, it is linked with the amount of the chlorophyll light harvesting protein complexes within the leaves [37]. From Figure 2c, the shape of the curves for leaves from the wild and the leaves from cultures is very similar - the progressions of the trends observed to be parallel. This trend could be interpreted in that the photosynthetic capacity of the plant is proportional to the preconditioning treatment of which the plant is exposed to. If the available light is lower, correspondingly, the curve will tend to be lowered as well or vice versa for higher light treatments until a cut-off minimum or maximum threshold. respectively. The P_{max} value is also lowered for treatments having lower light availability. This is an important phenomenon to note. For seagrasses living in a light attenuated and/or deeper natural environment, the maximal photosynthetic rate would still be lowered to a certain extent due to the lowered light availability despite the increased chlorophyll leaf content. From this study, the decrease is not very large if P_{max} was based on chlorophyll content solely (a reduction by 4.35% for *H. ovalis* leaves from cultures). But in terms of photosynthetic efficiency, the reduction in not very negligible. As can be seen from Table 1, the photosynthetic efficiency (α) between wild leaves and cultured leaves was lowered by as much as 11.38%.

This is, of course, if the basal parameter was analysed by leaf chlorophyll content only. The trends from Figures 2a (FW) and 2b (Area) presents a different predisposition compared to Figures 2c (Chl) . Although there is a tendency for the photosynthetic efficiency to be lowered in terms of chlorophyll content, but in terms of leaf weight and surface area, the trend is the opposite. The curve for leaves from cultures is elevated with respect to wild leaves. This is similar for P_{max} based on FW and Area. Combining the trends plotted from Figures 2a, b and c, it can be inferred that for leaves from cultures, the lowered P_{max} based on Chl was offset by a corresponding increase in P_{max} based on FW and Area. In terms of the photosynthetic capacity of the plant, the primary response to environmental light reduction would be an increase in the production of chlorophyll. A change in leaf morphology, i.e. larger leaf area and increased leaf weight is a secondary consequence. The likely reason for this is that the plant will tend to acclimatise itself, although to a limited extent, to the most similar levels in high or low light situations.

The results collated from this study implied that *H. ovalis* is very tolerant of low light conditions. This is very much the trend for other *Halophila* seagrasses (see Table 2). Based on the responses to the P-I curve, *H. ovalis* can be classified as a shade plant that has a high photosynthetic efficiency - the slope of α is steep, with a corresponding low I_c and I_k [38]. Other lower plants such as algae exhibit similar traits [32, 33]. Consequently, *H. ovalis*, and other *Halophila* are more similar in these physiological aspects to algae compared to terrestrial higher plants.

By observing the trends for the curves plotted, the disparity in the P-I relationships is likely due to the differences in light availability for proliferation between plants in the wild and plants in cultures.

Acknowledgements We would like to thank the Vice-Chancellor, Universiti Putra Malaysia, for encouragement and facilities. This research is funded by the Ministry of Science, Technology and Environment Malaysia, under the 'Intensification of Research in Priority Areas' programme entitled "Seagrass taxonomy, biology and habitat characteristics: EA-001-09-02-04-0679" and some financial and travel supports from Japan Society for the Promotion of Science (JSPS) are acknowledged.

REFERENCES

1. McMillan, C. (1983). Morphology of *Cymodocea serrulata* from Yap, Micronesia, kept under controlled light conditions. *Aquat. Bot.* **15**: 209 - 215.
2. Perez, M., Duarte, C.M., Romero, J., Sand-Jensen, K. and Alcoverro, T. (1994). Growth plasticity in *Cymodocea nodosa* stands: The importance of nutrient supply. *Aquat. Bot.* **47**: 249 - 264.
3. Abal, E., Loneragan, N., Bowen, P., Perry, C. and Udy, J. (1994). Physiological and morphological responses of the seagrass *Zostera capricorni* Aschers to light intensity. *J. Exp. Mar. Biol. Ecol.* **178**: 113 - 129.

4. Grice, A.M., Loneragan, N.R. and Dennison, W.C. (1996). Light intensity and the interactions between physiology, morphology and stable isotope ratios in five species of seagrass. *J. Exp. Mar. Biol. Ecol.* **195**: 91 - 110.
5. Jensen, S. and Bell, S., 2001. Seagrass growth and patch dynamics: Cross-scale morphological plasticity. *Pl. Ecol.* **155**: 201 - 217.
6. Ruiz, J.M. and Romero, J. (2001). Effects of *in situ* experimental shading on the Mediterranean seagrass *Posidonia oceanica. Mar. Ecol. Prog. Ser.* **215**: 107 - 120.
7. Lee K.-S. and Dunton, K.H. (1997). Effects of *in situ* light reduction on the maintenance, growth and partitioning of carbon resources in *Thalassia testudinum* Banks ex König. *J. Exp. Mar. Biol. Ecol.* **210**: 53 - 73.
8. Nelson, T. A. and Lee A. (2001). A manipulative experiment demonstrates that blooms of the macroalga *Ulvaria obscura* can reduce eelgrass shoot density. *Aquat. Bot.* **71**: 149 - 154.
9. Hemminga, M. and Duarte, C. (2000). *Seagrass Ecology*. Cambridge University Press, Cambridge, 298 pp.
10. Larkum, A.W.D., Roberts, G., Kuo, J. and Strother S. (1989). Gaseous movement in seagrasses. In: *Biology of Seagrasses* (eds. Larkum, A.W.D., McComb, A.J., Sheperd, S.A.). Elsevier, Amsterdam. pp. 686 - 722.
11. Beer, S. and Waisel, Y. (1982). Effects of light and pressure on photosynthesis in two seagrasses. *Aquat. Bot.* **13**: 331 - 337.
12. Evans, A.S., Webb, K.L. and Penhale, P.A. (1986). Photosynthetic temperature acclimation in two coexisting seagrasses, *Zostera marina* L. and *Ruppia maritima* L. *Aquat. Bot.* **24**: 185 - 197.
13. Abu Hena M.K., Misri K., Japar Sidik B., Hishamuddin O., Hidir H. (2001). Photosynthetic and respiration responses of dugong grass *Thalassia hemprichii* (Ehrenb) Aschers at Teluk Kemang seagrass bed, Malaysia. *Pakistan J. Biol. Sci.* **4**: 1487 - 1489.
14. Libes, M. (1986). Productivity-irradiance relationship of *Posidonia oceanica* and its epiphytes. *Aquat. Bot.* **26**: 285 - 306.
15. Hansen, J.W., Pedersen, A.U., Berntsen, J., Rønbøg, I.S., Hansen, L.S. and Lomstein B.A. (2000). Photosynthesis, respiration, and nitrogen uptake by different compartments of a *Zostera marina* community *Aquat. Bot.* **66**: 281 - 295.
16. Roberts, D.G. and Moriarty, D.J.W. (1987). Lacunal gas discharge as a measure of productivity in the seagrassses, *Zostera capricorni*, *Cymodocea serrulata* and *Syringodium isoetifolium*. *Aquat. Bot.* **28**: 143 - 160.
17. Erftemeijer, P.L.A., Osinga, R. and Mars, A.E. (1993). Primary production of seagrass beds in South Sulawesi (Indonesia): A comparison of habitats, methods and species. *Aquat. Bot.* **46**: 67 - 90.
18. Erftemeijer, P.L.A and Stapel, J. (1999). Primary production of deep-water *Halophila ovalis* meadows. *Aquat. Bot.* **65**: 71 - 82.
19. Rozaimi Jamaludin M., Japar Sidik B., Misri K., Hishammudin O. (2005). Shading responses of the seagrass *Halophila ovalis*: A preliminary comparison between cultures and naturally growing plants. *Malaysian J. Sci.* **24**: 23 - 29.
20. Japar Sidik B., Muta Harah Z., Aziz A., Mohd Pauzi A. (2001). Responses of *Halophila ovalis* and *Cymodocea serrulata* under the shade of *Enhalus acoroides*. In: *Aquatic Resource and Environmental Studies of the straits of Malacca: Current Research and Reviews.* (eds B. Japar Sidik, A. Arshad, S.G. Tan, S.K. Daud, H.A. Jambari, S. Sugiyama). Malacca Sraits Research and Development Centre (MASDEC). Univ. Putra Malaysia, Serdang, Malaysia. pp. 111-115.
21. Beer, S., Mtolera, M., Lyimo, T. and Bjork, T. (2006). The photosynthetic performance of the tropical seagrass *Halophila ovalis* in the upper intertidal. *Aquat. Bot.* **84**: 367 - 371.
22. Dennison, W.C. (1990). Seagrasses: Chlorophyll content. In: *Seagrass Research Methods* (ed. Phillips, R.C.). UNESCO, pp. 83 - 86.
23. Granger, S. and Izumi, H. (2001). Water quality measurement methods for seagrass habitats. In: *Global Seagrass Research Methods* (eds. Short, F.T., Coles R.G.). Elsevier, Amsterdam/New York, pp. 393 - 406.
24. Vermaat, J.E., Agawin, N.S.R., Duarte, C.M., Enriquez, S., Fortes, M.D., Marba, N., Uri J.S. and van Vierssen W. (1997). The capacity of seagrasses to survive increased turbidity and siltation: the significance of

growth form and light use. *Ambio*. **26**: 499 - 594.

25. Hillman, K., McComb, A.J. and Walker, D.I. (1995). The distribution, biomass, and primary production of the seagrass *Halophila ovalis* in the Swan/Canning Estuary, Western Australia. *Aquat. Bot.* **51**: 1 - 54.
26. Dawes, C.J., Lobban, C.S. and Tomasko, D.A. (1989). A comparison of the physiological ecology of the seagrasses *Halophila decipiens* Ostenfeld and *H. johnsonii* Eiseman from Florida. *Aquat. Bot.* **33**: 149 - 154.
27. Dawes, C., Chan, M., Chinn, R., Koch, E.W., Lazar, A. and Tomasko, D.A. (1987). Proximate composition, photosynthetic and respiratory responses of the seagrass *Halophila engelmanii* from Florida. *Aquat. Bot.* **27**: 195 - 201.
28. Dunton, K.H. and Tomasko, D.A. (1994). *In situ* photosynthesis in the seagrass *Halodule wrightii* in a hypersaline subtropical lagoon. *Mar. Ecol. Prog. Ser.* **107**: 281 - 293.
29. Herzka, S.K. and Dunton, K.H. (1997). Seasonal photosynhetic patterns of the seagrass *Thalassia testudinum* in the western Gulf of Mexico. *Mar. Ecol. Prog. Ser.* **152**: 103 - 17.
30. Drew, E.A. (1979). Physiological aspects of primary production in seagrasses. *Aquat. Bot.* 7: 139 - 150.
31. Masini, R.J. and Manning, C.R. (1997). The photosynthetic responses to irradiance and temperature of four meadow-forming seagrasses. *Aquat. Bot.* **58**: 21 - 36.
32. Myers, J. and Graham, J.-R. (1971). The photosynthetic unit in *Chlorella* measured by repititive short flashes. *Pl. Phys.* **71**: 440 - 442.
33. Ramus, J. (1981). The capture and transduction of light energy. In: *The Biology of Seaweeds* (eds. Lobban. C.S., Wynne, M.J.). Blackwell, Oxford. pp. 29 - 46.
34. Dennison, W.C. and Alberte, R.S. (1986). Photoadaptation and growth of *Zostera marina L.* (eelgrass) transplants along a depth gradient. *J. Exp. Mar. Biol. Ecol.* **98**: 265 - 282.
35. Pirc, H. (1986). Seasonal aspects of photosynthesis in *Posidonia oceanica*: Influence of depth, temperature and light intensity. *Aquat. Bot.* **26**: 203 - 212.
36. Vermaat, J.E. and Verhagen, F.C.A. (1996). Seasonal variation in the intertidal seagrass *Zostera noltii* Hornem.: Coupling demographic and physiological patterns. *Aquat. Bot.* **52**: 259 - 281.
37. Pearcy, R.W. (1998). Acclimation to sun and shade. In: *Photosynthesis: A comprehensive treatise* (ed. Raghavendra A.S.). Cambridge University Press, New York. pp. 250 - 263.
38. Dawes, C.J. (1998). *Marine Botany*, John Wiley, New York, 628 pp.