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Seasonality and depth zonation of intertidal *Halophila ovalis* and *Zostera japonica* in Ha Long Bay (northern Vietnam)

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Abstract

Seasonality and depth zonation of two common intertidal northern Vietnamese seagrass species, *Halophila ovalis* and *Zostera japonica*, were investigated in a 50 m long intertidal transect from 1999 till 2001. *Z. japonica* occurred in the middle intertidal (1.2 m above mean chart datum to mean sea level at 2.1 m above chart datum). *H. ovalis* occurred deeper in the intertidal (between 1.0 and 1.4 m above chart datum). Based on seasonally different sensitivity to reciprocal transplants, it is concluded that this zonation is most likely due to different tolerances to low light availability (less in *Z. japonica*) and desiccation (less in *H. ovalis*). Both species reached maximum shoot densities in September–October with total biomasses around 70 g DW m⁻². Flowering in *Z. japonica* occurred in April only, whilst *H. ovalis* flowered in November after the rainy season and again in April. Seasonality in density and biomass was apparent and similar between the two species. It was

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largely coupled to the rainy season, which brings spates of turbid water during May–August, thereby possibly limiting light availability and hence growth.

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1. Introduction

The seagrass beds of subtropical northern Vietnam are in a transition zone between those of temperate and northern Asian shores where several *Zostera* species prevail and those of tropical southern Vietnam with the high species richness of the Indo-Pacific flora (Den Hartog, 1970; Phillips and Menez, 1988). Northern *Zostera japonica* extends down to the latitude of Hue, and tropical *Halophila ovalis* occurs northward up to Ha Long Bay (Tien, 1998). The distribution ranges of these two small, fast-growing species, which occupy similar niches in intertidal stands, overlap in the Ha Long area (Tien, 1998).

Particularly in this transition region, the ecology of the co-occurring seagrass species has received little attention, with only floristic surveys available (Tien, 1998). In the central parts of its distribution, *Z. japonica* occurs mainly intertidally (Harrison, 1982a; Baldwin and James, 1994; Lee, 1997), whereas *H. ovalis* may occur both intertidally and permanently submerged (Duarte, 1991a; Hillman et al., 1995; Vermaat et al., 1995; Longstaff and Dennison, 1999; Nakaoka and Aioi, 1999). Therefore, at present it is unclear whether these two species will co-occur in mixed stands or segregate in depth zones. Furthermore, the contrasting distribution ranges suggest different habitat requirements, which in turn may cause different responses to seasonal variation in forcing environmental factors.

The present study examined depth distribution and seasonality in growth and density of these two co-occurring seagrass species. The results from in situ transects and reciprocal transplantation experiments demonstrate a partial depth segregation and contrasting growth patterns.

2. Materials and methods

2.1. Study site

The main study site was located at Gia Luan (northern Cat Ba Island, Ha Long Bay, northern Vietnam, 20°51.02'N–106°58.96'E). The site is one of many relatively undisturbed small bays in the area with mangrove stands, intertidal seagrass beds and mudflats. The dominant seagrasses in Gia Luan are *H. ovalis* and *Z. japonica*. Additional data were collected from Dau Moi (a cove on the mainland coast east of Ha Long City, 25 km northeast of Gia Luan) in April 1999, where the same two seagrass species occurred together with scattered *Halophila decipiens*.

Tidal exposure data were extracted from tidal tables published for Hong Gai (western Ha Long City, Quang Ninh province) by the Vietnamese Naval High Command (Marine

Hydrometeo Centre, 1998). Salinity, water temperature and rainfall data were taken for a permanent monitoring station at nearby Ha Long City operated by the Hai Phong Institute of Oceanology (Hoi and Dieu, 1995). Water turbidity was measured in adjacent deeper water during the sampling visits with a Secchi disk.

2.2. Seasonality of seagrass density and growth

Monitoring of plant biomass was accomplished by collecting all seagrass material within four replicate quadrats (20 cm × 20 cm) at 10 m intervals along a transect of 50 m length perpendicular to the water line. In April 1999, two transects were sampled in Gia Luan, and one in Dau Moi. Seagrass sampling continued monthly on the second transect at Gia Luan until May 2000. The samples were washed to remove sediment, and brought to the laboratory for further measurements. In the laboratory, the number of shoots, apices and flowers, the length of sheaths or petioles, leaves and rhizome internode segments were recorded. Then the material was separated into above-ground (vegetative shoots and flowering shoots) and below-ground fractions (rhizomes and roots), dried at 105 °C during 24 h, and subsequently weighed.

With the same frequency, 20 rhizome apices were tagged by placing a marker at the third shoot from the rhizome apex. Leaf lengths of the third and second shoot from the apex were measured. These marked plants were re-collected after 7 days. *Z. japonica* was marked in the upper part of the transect and *H. ovalis* in the lower part, in accordance with their abundance. After return to the laboratory, each marked rhizome piece was measured individually, leading to observations of rhizome material produced and number of shoots produced per rhizome apex. For the newly produced shoots, the number of leaves and the biomass were quantified. Rhizome elongation was estimated from the new internodes grown at the apex, and leaf elongation from the difference in leaf lengths on the third and second shoot from the apex. Above-ground productivity was estimated from monthly data on shoot density and leaf growth data in *Z. japonica* and from rhizome elongation, internode and apex density as well as shoot biomass in *H. ovalis*. The latter is based on the assumption that each new internode bears an apical shoot.

2.3. Reciprocal transplantation experiments

The experiments were carried out four times from April 2000 to February 2001. In each experiment, we excavated three 20 cm × 20 cm quadrats of *Z. japonica* from monospecific stands in the higher intertidal, and transplanted these to three holes excavated in the *H. ovalis* zone. Three additional quadrats were excavated in *Z. japonica*, but these were placed back again to estimate any effects that excavation might have had on the plants (procedural controls). For *H. ovalis*, we proceeded in the same manner: 20 cm × 20 cm sods were taken from monospecific stands in the lower intertidal and placed in the *Z. japonica* zone; procedural controls were established as well. After 1 month, all quadrats were harvested and taken to the laboratory for further measurements as described above. One month was taken as the experimental period, because responses should be sufficiently clear after such a period for these comparatively short-lived species (Duarte, 1991b).

Repeated examination of the sods in situ was too difficult logistically dictating analysis in the laboratory.

3. Results

Seasonal variation in rainfall was profound, with the maximum occurring during the rainy monsoon season of May–September (Fig. 1). During this period, salinity in Ha Long

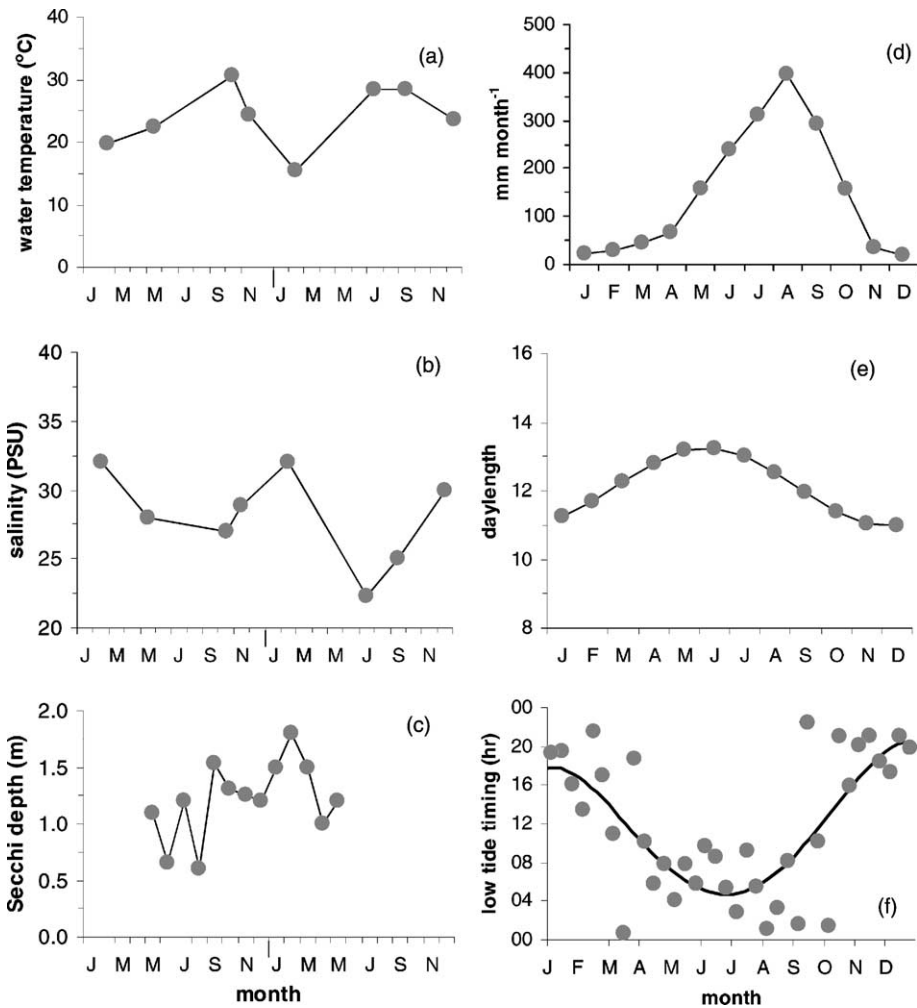


Fig. 1. Seasonality in environmental factors: (a) water temperature (°C); (b) surface water salinity (psu); (c) Secchi disk transparency (m); (d) rainfall (mm per month); (e) day length (h); and (f) timing of low tide during the day. Note difference in horizontal axis between (a)–(c) and (d)–(f). Data in (a)–(c) are from in situ observations at Gia Luan, those in (e)–(f) are from other sources (see Section 2).

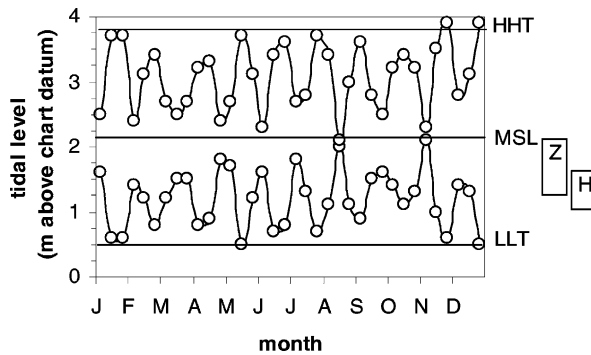


Fig. 2. Low tide (lower curve) and high tide water levels (upper curve) at Ha Long City in 1999 according to Marine Hydrometeo Centre (1998). Also presented are the highest high tide (HHT) and lowest low tide (LLT) reported, as well as the median taken to represent mean sea level (MSL). The rectangles to the right depict the tidal depth range of *Z. japonica* (Z) and *H. ovalis* (H).

Bay declined from 32 to 22 psu in July 2000, and water turbidity fluctuated with periods of quite low Secchi depth (Fig. 1). Water temperature and day length followed the regular northern Hemisphere cycle with lowest temperatures occurring in January–February. The tidal regime is diurnal, leading to a distinct periodicity in low tide exposure during the day: from May to August the low tides occurred in the early morning, but from September till April in 2000 this was in the late afternoons (Fig. 1). The spring-neap cycle caused a considerable fluctuation in tidal amplitudes, ranging between less than 25 cm to more than 375 cm (Fig. 2). This cycle led to a pronounced periodicity in low tide exposure: during 1 year *H. ovalis* experienced nine emergence periods of about half-a-month alternating with periods without tidal emergence (Fig. 2), whereas *Z. japonica* generally experienced exposure every day.

Depth zonation of the seagrasses was distinct, but segregation was only partial (Figs. 2 and 3): *Z. japonica* occupied the middle intertidal (1.2–2.1 m above chart datum) up to the edge of the mangroves (*Kandelia candel* and *Rhizophora stylosa*) and *H. ovalis* the lower end (1.0–1.4 m).

Monthly variation in shoot density and biomass was substantial (Fig. 4). Density of both species increased rapidly by the end of the monsoon season. Subsequently, *H. ovalis* density declined after October 1999, coinciding with a maximum in flowering (Fig. 4). A second flowering event occurred in April 2000; hence this species did neither flower during the rainy season, nor during the colder January winter period. *Z. japonica* did not show such a sudden decline in density as *H. ovalis*, and density remained similar until February, soon after which flowering shoots were observed. During the flowering season, the leaf area of *Z. japonica* increased (Fig. 4). In *H. ovalis*, between 13 and 21% of the shoots were reproductive during peak flowering seasons, whereas this was 9% at most for *Z. japonica*. Leaf and rhizome elongation rates of both species showed a clear seasonal pattern, with a maximum in October (Fig. 4).

Stepwise multiple regressions of seagrass shoot density and biomass with water temperature, rainfall, salinity, daylength and Secchi depth led to a limited number of significant

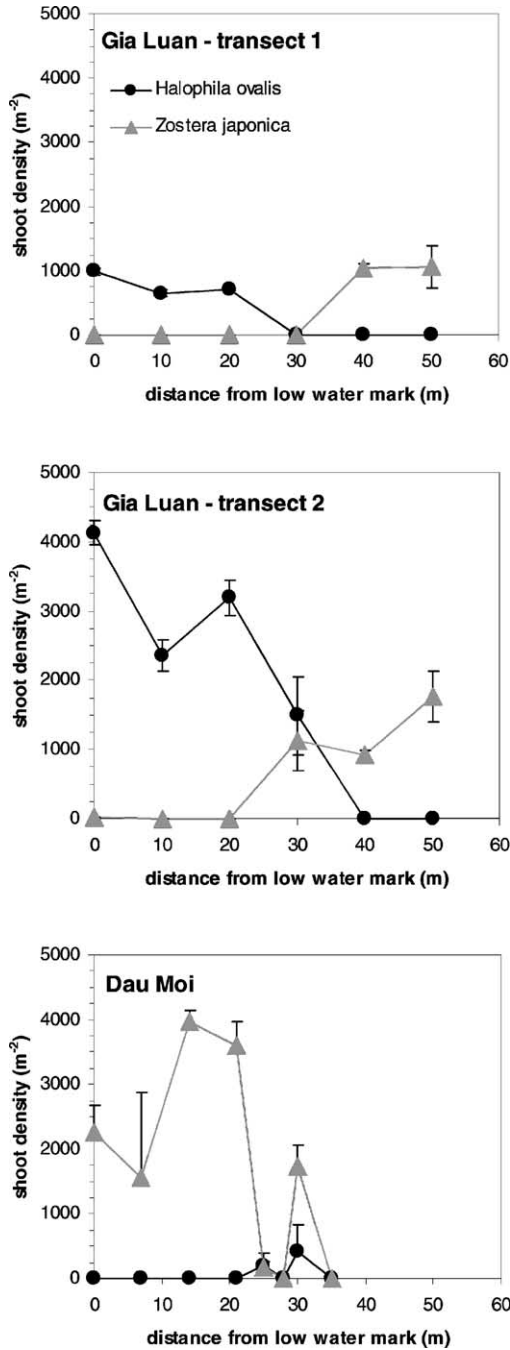


Fig. 3. Depth zonation of shoot density of *H. ovalis* and *Z. japonica* along the transect in Gia Luan (two transects separated 50 m) and Dau Moi, April 1999. Presented are mean ± 1 S.E.

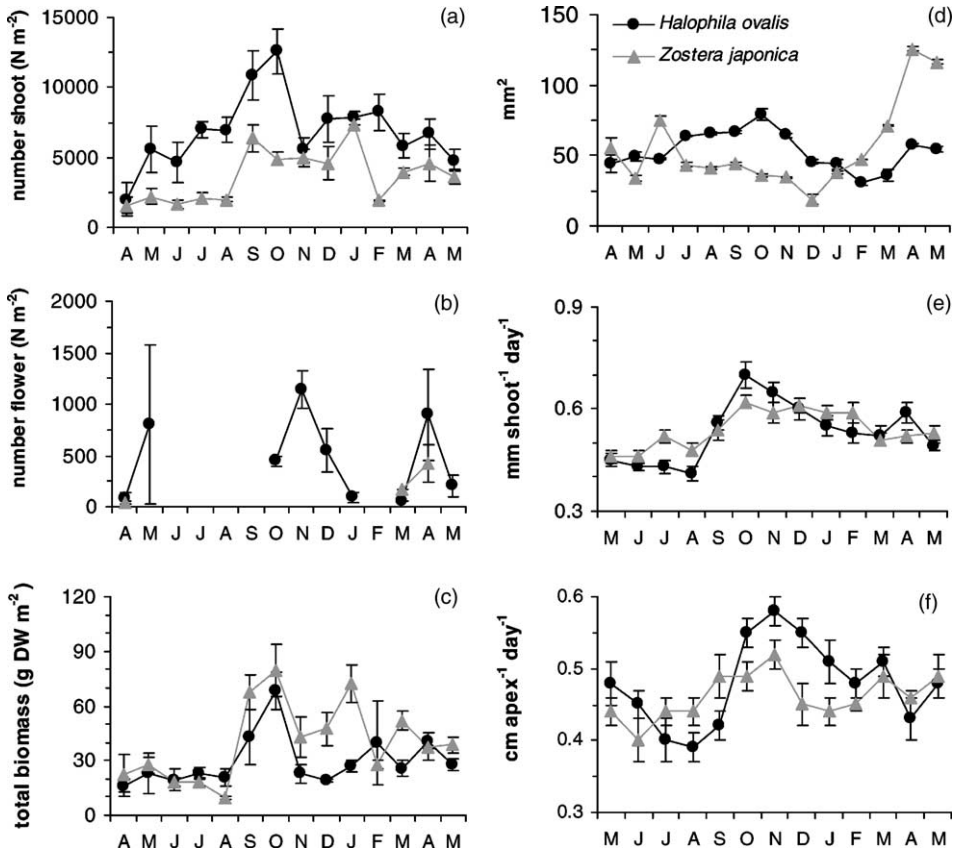


Fig. 4. Seasonality for both seagrass species *H. ovalis* and *Z. japonica* in (a) shoot density; (b) flowering shoot density; (c) total biomass; (d) single-sided shoot leaf area; (e) leaf elongation; (f) rhizome elongation. Data are presented as mean ± 1 S.E.

linear correlations. In *Z. japonica*, both shoot density and total biomass were negatively correlated with day length ($r^2 = 0.38$, $P < 0.025$). In *H. ovalis*, shoot density was negatively correlated to Secchi depth ($r^2 = 0.31$, $P = 0.05$).

Annual above-ground productivity estimates amounted to 140 and 410 g DW m⁻² per year, for *Z. japonica* and *H. ovalis*, respectively. Annual rhizome elongation rates were 168 ± 3 and 175 ± 7 cm per apex per year, respectively.

In the transplantation experiments, the patterns in shoot density and biomass were not completely similar (Table 1, Fig. 5): significant differences existed between the species in density, but not in biomass. Also, the difference between months was significant for density, but not for biomass. The significant species × month interaction for biomass suggests that *Z. japonica* was more strongly affected in some months than *H. ovalis*, notably in the period November–December (Fig. 5). The transplantation of each species into the zone of the other led to a significant reduction in shoot density as well as in total biomass. The ratio of above-to

Table 1

Reciprocal transplantation experiments with *H. ovalis* and *Z. japonica*: three-way ANOVA for number of shoots, total biomass, the ratio of above- to below-ground biomass and shoot leaf area with species, month and treatment (transplanted to the other zone or procedural control) as factors and only the species \times month interaction (others were not significant)

Parameter	Species		Month		Treatment		Species \times month	
	TSS (%)	<i>P</i>	TSS (%)	<i>P</i>	TSS (%)	<i>P</i>	TSS (%)	<i>P</i>
Shoot density (m^{-2})	11	0.001	44	0.001	7	0.001	12	0.003
Total biomass (g DW m^{-2})	5	0.53	3	0.439	17	0.001	23	0.002
Above–below	14	0.003	0	0.631	0	0.93	30	0.001
Shoot leaf area (cm^2 per shoot)	2	0.128	21	0.001	0	0.406	51	0.001

Presented is the percentage of variance explained by a factor (TSS = $(100 \times \text{factor SS})/\text{total SS}$), and the level of significance (*P*).

below-ground biomass was significantly higher in *H. ovalis* than in *Z. japonica* and also the species \times month interaction was significant. Shoot leaf area did not differ among species, but showed a significant seasonality, just as in the non-experimental transects. Overall, density and biomass in the experimental controls were similar to the values found in the transects.

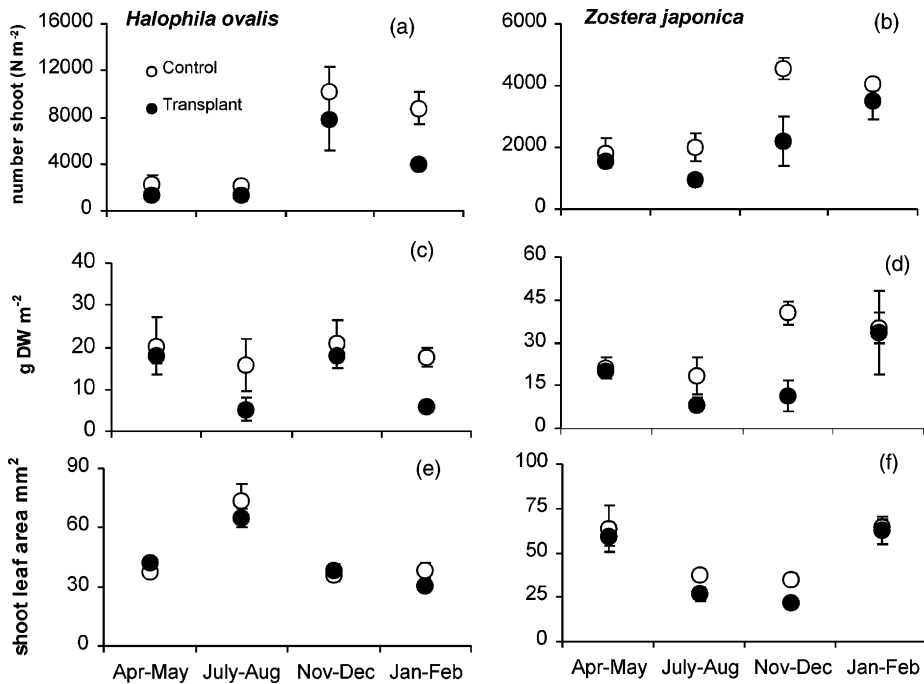


Fig. 5. Effects of reciprocal transplantation experiments carried out in four periods on (a) shoot density of *H. ovalis*; (b) shoot density of *Z. japonica*; (c) total biomass of *H. ovalis*; (d) total biomass of *Z. japonica*; (e) single-sided shoot leaf area of *H. ovalis*; (f) single-sided shoot leaf area of *Z. japonica*. Data are presented as mean ± 1 S.E.; $n = 3$.

4. Discussion

The seasonality of these two co-occurring seagrass species was found to be distinct (Fig. 4), and growth, shoot density and biomass tended to increase to maxima in the months after the rainy season, i.e. September and October. This pattern is in contrast to the seasonality observed for seagrasses at higher latitudes, where the solar daylength cycle governs most of the seasonality through light availability (for *Z. japonica*: Harrison, 1982b; for other species: Duarte, 1989; Marbà et al., 1996; Vermaat and Verhagen, 1996; Duarte and Chiscano, 1999). The seasonal pattern in shoot size and flowering differed between species. *Z. japonica* only flowered in spring, whilst *H. ovalis* flowered twice: in November after the rainy season and in April, together with *Z. japonica*. Also, a higher proportion of the shoots engaged in flowering in *H. ovalis*. In *H. ovalis*, shoot density, biomass as well as rhizome elongation were comparable to the range reported elsewhere (Hillman et al., 1995; Vermaat et al., 1995; Nakaoka and Aioi, 1999). For *Z. japonica*, density and biomass were high compared to published values (e.g. 20–40 g DW m⁻²; Harrison, 1982b; Baldwin and James, 1994; Lee, 1997). In situ rhizome elongation data have not been published so far, but can be roughly deduced from Bigley and Harrison (1986) using a median internode length of 1 cm and approximately 24 leaves as well as internodes produced per apex in a year (a plastochron interval of about 14 days; Hemminga and Duarte, 2000): this rough estimate of 24 cm per apex per year is not far from our value of 17 cm per apex per year. Our estimates of annual above-ground productivity were comparatively low (cf. Duarte, 1989, 1991b; Duarte and Chiscano, 1999; Hillman et al., 1995; Vermaat et al., 1997; Marbà and Duarte, 1998; Nakaoka and Aioi, 1999).

The observed depth zonation of the two species suggests a difference in habitat requirements. A similar, partial segregation was reported by Ramirez-Garcia et al. (1998) for two *Phyllospadix* species along the Pacific coast of Mexico, and a different sensitivity to desiccation at low tide was suggested as causal factor. In the present study, shoot density of both species declined after transplantation into the zone of the other species. Remarkably, we did not find any effects on individual shoot size. Apparently the potentially present plasticity in shoot size in both species (Harrison, 1982b; Hemminga and Duarte, 2000) was not induced by our experimental transplantation. The occurrence of *Z. japonica* in the higher intertidal is in agreement with observations by Harrison (1982a,b). Probably, this species has equally high light requirements as *Zostera noltii* (Vermaat and Verhagen, 1996), which is closely related (Bigley and Barreca, 1982). This is consistent with its low biomass in the turbid rainy season and the stronger effect of transplantation during the rains and fall. *H. ovalis*, on the other hand, is probably highly flexible in its acclimation to light, since it is reported from intertidal, from shallow subtidal as an understory species, as well as from very deep environments (Vermaat et al., 1995; Erftemeijer and Stapel, 1999; Hemminga and Duarte, 2000). The stronger effect of transplantation in *H. ovalis* outside the rainy season may have several explanations: low tide exposure during day time (as in Ramirez-Garcia et al., 1998), but also the low temperatures in January (growth is reduced strongly below 20 °C; Hillman et al., 1995) may have been stressful. During the rainy season, low salinities may have been adverse although we did not observe the lower limit of 20 psu suggested by Hillman et al. (1995). It should be noted here that the reduced vitality of the transplants could not have been due to a local sediment-related effect, since the plants were transplanted as whole,

intact sods. Also, shoot mortality was significant, but after each experiment still considerable numbers of shoots survived (40% or more, Fig. 5).

Flowering frequency was comparatively low in both species. In the tidally more exposed station at Dau Moi, we observed that *Z. japonica* produced higher quantities of flowering shoots in April 1999 (up to 13%, whilst the species hardly flowered in Gia Luan by that time), after which massive shoot mortality occurred in June. This conforms with the observations of Harrison (1982b) on the Canadian Pacific coast, who also found an annual strategy in populations of *Z. japonica* that were exposed to longer tidal emergence. In Canada, however, mortality occurred in September, at the onset of winter, and not before the rainy season.

In short, we demonstrated that depth zonation of the two commonest intertidal northern Vietnamese seagrass species is likely due to different habitat requirements. Seasonality in density and biomass was apparently similar between the two species, and minima occurred largely in the rainy season, which brings spates of turbid water during May–August, thereby possibly limiting light availability and hence growth.

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References

- Baldwin, J.R., James, R.L., 1994. Expansion of seagrass habitat by the exotic *Zostera japonica*, and its use by dabbling ducks and brant in boundary, British Columbia. *Mar. Ecol. Progr. Ser.* 103, 119–127.
- Bigley, R.E., Barreca, J.L., 1982. Evidence for synonymizing *Zostera americana* den Hartog with *Zostera japonica*. *Aschers. Graebn. Aquat. Bot.* 14, 349–356.
- Bigley, R.E., Harrison, P.G., 1986. Shoot demography and morphology of *Zostera japonica* and *Ruppia maritima* from British Columbia, Canada. *Aquat. Bot.* 24, 69–82.
- Den Hartog, C., 1970. *The Seagrasses of the World*. North-Holland, Amsterdam.
- Duarte, C.M., 1989. Temporal biomass variability and production/biomass relationships of seagrass communities. *Mar. Ecol. Progr. Ser.* 51, 269–276.
- Duarte, C.M., 1991a. Seagrass depth limits. *Aquat. Bot.* 40, 363–377.
- Duarte, C.M., 1991b. Allometric scaling of seagrass form and productivity. *Mar. Ecol. Progr. Ser.* 77, 289–300.
- Duarte, C.M., Chiscano, C.L., 1999. Seagrass biomass and production: a reassessment. *Aquat. Bot.* 65, 159–174.
- Erfemeijer, P.L.A., Stapel, J., 1999. Primary production of deep-water *Halophila ovalis* meadows. *Aquat. Bot.* 65, 71–82.
- Harrison, P.G., 1982a. Comparative growth of *Zostera japonica* As. and *Zostera marina* L. under simulated intertidal and subtidal conditions. *Aquat. Bot.* 14, 373–379.
- Harrison, P.G., 1982b. Seasonal and year-to-year variations in mixed intertidal populations of *Zostera japonica* Aschers and graebn. and *Ruppia maritima* L. s.l. *Aquat. Bot.* 14, 357–371.
- Hemminga, M.A., Duarte, C.M., 2000. *Seagrass Ecology*. Cambridge University Press, Cambridge.
- Hillman, K., McComb, A.J., 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.
- Hoi, N.C., Dieu, L.V., 1995. Investigation and assessment of environment in the north of Vietnam. IDRC project final report.
- Lee, S.Y., 1997. Annual cycle of biomass of a threatened population of the intertidal seagrass *Zostera japonica* in Hong Kong. *Mar. Biol.* 129, 183–193.

- Longstaff, B.J., Dennison, W.C., 1999. Seagrass survival during pulsed turbidity events: the effects of light deprivation on the seagrasses *Halodule pinifolia* and *Halophila ovalis*. *Aquat. Bot.* 65, 105–121.
- Marbà, N., Duarte, C.M., 1998. Rhizome elongation and seagrass clonal growth. *Mar. Ecol. Progr. Ser.* 174, 269–280.
- Marbà, N., Cebrián, J., Enríquez, S., Duarte, C.M., 1996. Growth patterns of western Mediterranean seagrasses: species-specific responses to seasonal forcing. *Mar. Ecol. Progr. Ser.* 133, 203–215.
- Marine Hydrometeo Centre, 1998. Tidal Tables, vol. 1. Hanoi, Vietnam, 177 pp.
- Nakaoka, M., Aioi, K., 1999. Growth of seagrass *Halophila ovalis* at dugong trails compared to existing within-patch variation in a Thailand intertidal flat. *Mar. Ecol. Progr. Ser.* 184, 97–103.
- Phillips, R.C., Menez, E.G., 1988. Seagrasses. Publications of the Smithsonian Institution, No. 34. Washington, DC, 105 pp.
- Ramirez-García, P., Lot, A., Duarte, C.M., Terrados, J., Agawin, N., 1998. Bathymetric distribution, biomass and growth dynamics of intertidal *Phyllospadix scouleri* and *Phyllospadix torreyi* in Baja California (Mexico). *Mar. Ecol. Progr. Ser.* 173, 13–23.
- Tien, N.V., 1998. Composition and distribution of seagrass in Quang Ninh, Vietnam. *Resources Mar. Environ.* 5, 36–42.
- Vermaat, J.E., Verhagen, F.C.A., 1996. Seasonal variation in the intertidal seagrass *Zostera noltii* Hornem.: coupling demographic and physiological patterns. *Aquat. Bot.* 52, 259–281.
- Vermaat, J.E., Fortes, M.D., Agawin, N., Duarte, C.M., Marbà, N., Uri, J., 1995. Meadow maintenance, growth and productivity in a mixed Philippine seagrass bed. *Mar. Ecol. Progr. Ser.* 124, 215–225.
- Vermaat, J.E., Agawin, N., Duarte, C.M., Enríquez, S., Fortes, M.D., Marbà, N., Uri, J., 1997. The capacity of seagrasses to survive eutrophication and siltation, the significance of growth form and light use. *Ambiology* 26, 409–504.