



# Testing the effect–response framework: key response and effect traits determining above-ground biomass of salt marshes

Vanessa Minden & Michael Kleyer

## Keywords

Allometric relationships; Effect trait; Path analysis; Response trait; Structural equation modelling; Trade-off

## Abbreviations

SLA, specific leaf area; MAOS, morphological adaptations to osmotic stress; AGB, above-ground biomass

## Nomenclature

SynBioSys Species Checklist (2010)

Received 10 September 2010

Accepted 02 February 2011

Co-ordinating Editor: Lindsay Turnbull

**Minden, V.** (Corresponding author, [vanessa.minden@uni-oldenburg.de](mailto:vanessa.minden@uni-oldenburg.de)) &

**Kleyer, M.** ([michael.kleyer@uni-oldenburg.de](mailto:michael.kleyer@uni-oldenburg.de)): Landscape Ecology Group, Institute of Biology and Environmental Sciences, University of Oldenburg, D-26111 Oldenburg, Germany

## Abstract

**Question:** How do species traits respond to environmental conditions and what is their effect on ecosystem properties?

**Location:** Salt marshes, Northwest Germany.

**Methods:** On 113 plots along the German mainland coast and on one island, we measured environmental parameters (soil nutrient content, inundation frequency, groundwater level and salinity), collected traits from 242 individuals (specific leaf area [SLA], whole plant C:N ratio, and dry weights of plant organs) and sampled above-ground biomass as an ecosystem property. We constructed a path model combining environmental parameters, functional traits at community level and above-ground biomass, which was tested against a dependence model using path analysis; model fit was evaluated by structural equation modelling (SEM).

**Results:** The final model showed good consistency with the data and highlights the major role of groundwater level, salinity and nutrient availability as the most important factors influencing biomass allocation in salt marshes. Above-ground living biomass was mostly determined by stem biomass, which was mediated through an allometric allocation of biomass to all other plant organs, including leaf mass. C:N ratio and SLA were the major drivers for dead biomass.

**Conclusion:** We emphasize an indirect link between standing biomass and environmental conditions and recognize stem biomass, plant C:N ratio and SLA as keystone markers of species functioning in determining the relationship between environment and ecosystem properties.

## Introduction

In recent years, there has been growing consensus that the use of functional traits of plants, rather than taxonomic specification, can assist in explaining vegetation-related ecosystem properties and functions (Hooper et al. 2005). Lavorel & Garnier (2002) developed a conceptual framework that focuses on the prediction of changes in ecosystem processes using plant functional traits. It distinguishes between functional response traits (set of traits associated with the response of plants to environmental factors, such as allocation of biomass as a response to nutrient availability, Gitay & Noble 1997; Walker et al. 1999) and functional effect traits (set of traits related to the effect of plants on the environment, such as the effect of plant traits on the decomposability of litter, Díaz &

Cabido 2001). However, response and effect traits sometimes overlap, i.e. traits can explain both plant responses to environmental conditions and effects on the ecosystem (Lavorel & Garnier 2002). This is said to be particularly the case in the use of resources (Díaz & Cabido 2001) and in primary productivity (Lavorel & Garnier 2002). Therefore, Suding et al. (2008) attempted to simultaneously identify traits that respond to environmental gradients and traits affecting ecosystem properties. In our paper, we apply this approach to salt marshes of Northwest Germany by linking plant functional traits with abiotic conditions and ecosystem properties, which are live and dead above-ground biomass (AGB).

AGB is a major component of ecosystem functioning, being a temporary pool of fixed carbon, and providing the basis for agricultural exploitation by cattle grazing and

mowing. AGB of salt marshes is also of major importance for nature conservation, e.g. by providing feeding grounds for many bird species, especially for migratory geese (Bakker et al. 2005; Blew et al. 2005). Various studies have focused on the production of biomass in salt marshes (Janiesch 1991; Bakker et al. 1993); however, to our knowledge, there is still a lack of studies on linkages between environmental conditions, plant functional traits and biomass production. In particular, studies that merge the field of botanical scaling (Enquist & Niklas 2002) with scaling from environment through communities and ecosystem properties (Naeem & Wright 2003; Suding et al. 2008) are absent. Salt marshes are particularly suited for this purpose because their plant species pool is relatively small. Therefore, a single study can quantify species biomass allocation patterns, environmental conditions and ecosystem properties.

The main emphases of this paper are the assignment of the effect–response framework to salt marsh habitats, as well as the exploration of allometric and non-allometric relationships among plant traits, in order to understand the functional relationships that determine ecosystem properties. According to de Bello et al. (2010), it is necessary to identify key characteristics and mechanisms by which organisms affect ecosystem properties in order to promote the sustainable use of ecosystems. In this study, we address the questions of how species traits respond to environmental conditions of salt marshes and devise trait interactions and trade-offs between functional traits of salt marsh plants. Finally, we point out effect traits that determine production of standing biomass in salt marshes and give suggestions for the application of our results to theoretical modelling and biological conservation. We hypothesize that (i) trade-offs and allometries in biomass allocation determine the relationship between environmental conditions and community standing biomass in the response–effect framework; (ii) resources and abiotic stress factors such as salt and inundation are the primary agents determining response traits (Lavorel & Garnier 2002); and (iii) we expect a small set of keystone traits will define the ecosystem properties.

To specify our assumptions, we constructed a conceptual model based on *a priori* knowledge, which was tested against a dependence model using path analysis and structural equation modelling (Grace & Pugsek 1997). These techniques allow for the evaluation of alternative models, partitioning of direct and indirect effects and test for model fit (Mitchell 1992; Grace & Pugsek 1998). Furthermore, to explore how scaling relationships translate from the species to the community level, we determined bivariate scaling relationships between biomass allocated to roots and rhizomes, leaves, stems and diaspores at the level of individual plants.

## The hypothesised model: Linkages between traits

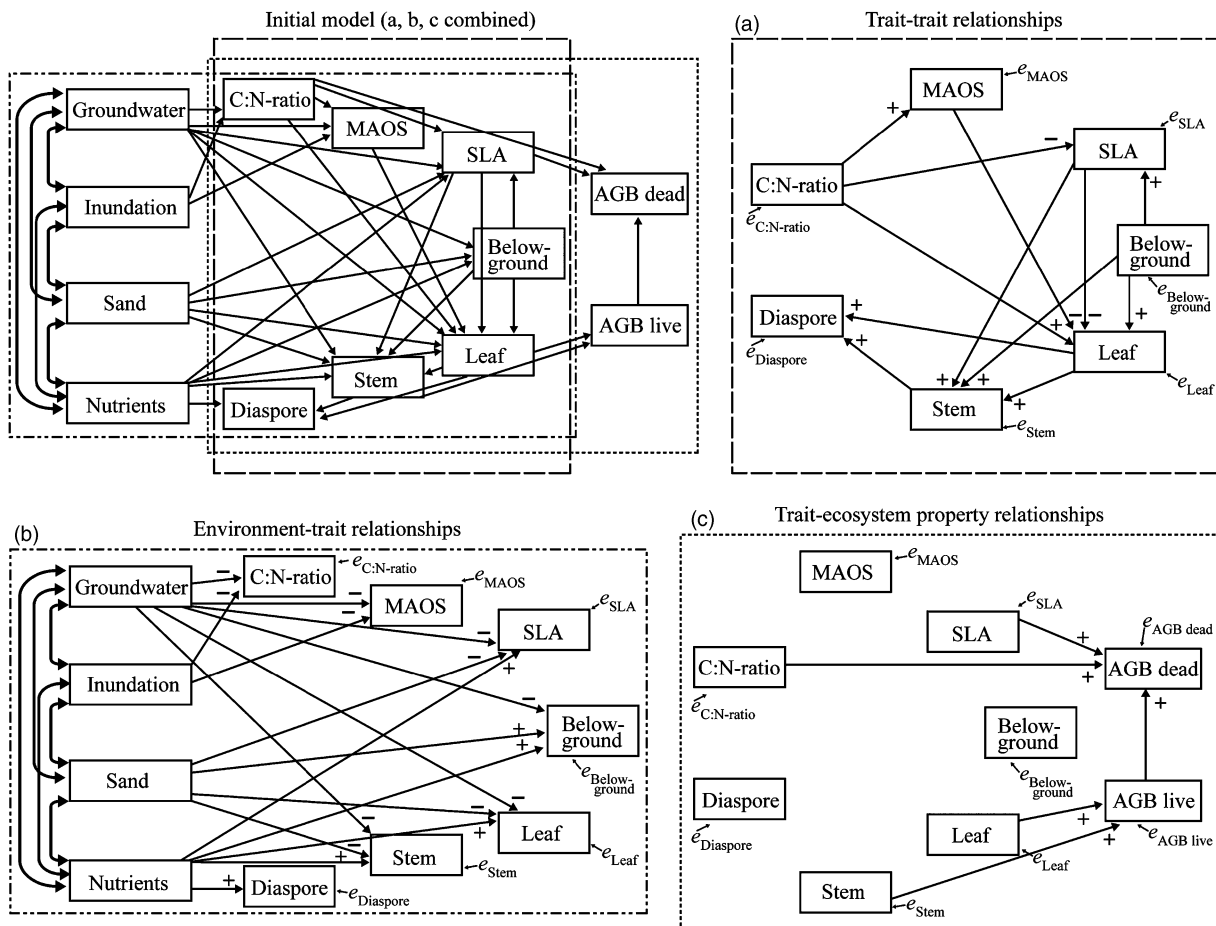
We begin the evaluation of the hypothesised model with linkages between plant traits because these determine the biological functioning of plants (Fig. 1a). Thereafter, we connect the traits with the environmental variables (Fig. 1b) and biomass variables (Fig. 1c).

Allocation theory assumes that organisms have a limited supply of resources that they have to allocate amongst their functions, broadly defined as growth, maintenance, storage and reproduction (Bazzaz 1997). Two different perspectives can be found in the literature: ‘partitioning’ and ‘allometry’ (Weiner et al. 2009). The partitioning perspective is interested in shifts in allocation patterns among plant structures and functions on environmental gradients, expressed as ratios such as root:shoot (Obeso 2002). Ratio-based biomass partitioning measures are size-independent, i.e. a large plant may exhibit the same root:shoot ratio as a small plant. The allometry perspective emphasizes variation in size and states that scaling relations among standing leaf, stem and root biomass are generally positive, e.g. that root biomass increases with leaf and stem biomass (Enquist & Niklas 2002). As we are interested in understanding the variation of a size-dependent ecosystem variable in relation to plant biomass allocation patterns and environmental conditions, we adopted the allometry perspective and quantified the dry weight of leaves, stems, diaspores and below-ground organs (i.e. root and rhizome mass).

Leaves (and to a lesser extent, stems) provide the plant with assimilation products; hence we expect a positive relationship between leaf biomass, on the one hand, and stem and diaspore biomass, on the other hand (Fig. 1a, Enquist & Niklas 2002; Niklas & Enquist 2003; Weiner et al. 2009).

We expect a negative relationship from C:N ratio to SLA and a positive relationship from C:N ratio to leaf mass (Wright et al. 2004). In contrast, we expect a negative relationship from SLA to leaf and a positive relationship to stem biomass, as we expect leaf mass of fast-growing species with high SLA to be lower than that of species that retain their nutrients and biomass (species with low SLA and high C:N-ratios). Furthermore, plants with high allocation to stem biomass are able to support high diaspore biomass (Thompson et al. 1991), thus we expect a positive relationship between these traits.

Below-ground mass provides the basis for above-ground growth, thus we draw a positive relationship between this trait and SLA, leaf and stem biomass. Three general strategies to cope with salt stress have been identified: (i) exclusion of salt ions, (ii) dilution of cell sap, and (iii) morphological adaptations to osmotic stress. The first strategy refers to species having structural and



**Fig. 1.** Hypothesized model of relationships between environmental parameters, traits and ecosystem property (Initial model, upper left). From the initial model, relationships between traits (a, upper right), environment–trait relationships (b, bottom left) and trait–ecosystem property relationships (c, bottom right) were magnified for a better overview. Allometric relationships are indicated by '+', negative relationships by '-', see also text above. Single-headed arrows represent relationships; double-headed arrows represent free correlations. Residual error variables ( $e_x$ ) represent effects of unexplained causes (omitted from initial model). For names and abbreviations see Table 1.

functional adaptations that reduce ion uptake through the root (Kinzel 1982), excrete salt ions via glands and bladders, or continuously renew the basal leaves (Schirmer & Breckle 1982; Van Diggelen et al. 1986). Salt-diluting species accomplish dilution of cell sap via enlargement of mesophyll cells, which results in succulent growth (Kinzel 1982). Species that show morphological adaptations to osmotic stress roll in their leaves to decrease the transpiring area or cover their leaves with hairs to attenuate incoming solar radiation (Rozema et al. 1985), which ultimately minimizes the transpiration rate and intake of saline water. Some species of the salt marsh show a combination of several of these strategies, whereas others only exhibit a single adaptation. For our model, we classified the salt marsh species into two groups: those featuring excretion and dilution, and those featuring morphological adaptations to osmotic stress (MAOS). The latter applies to species of the upper salt marsh

(*Elymus athericus*, *Festuca rubra* and *Artemisia maritima*), which do not show any other adaptations to salt stress. It could be assumed that reduced leaf mass could provide an advantage to those species, as the transpiring area is reduced. We thus expect a negative relationship between MAOS and leaf biomass. However, we expect a positive relationship between C:N ratio of the whole plant and MAOS, as the latter often is accompanied by xeromorphic tissue (Rozema et al. 1985).

#### The hypothesized model: linkages between environment, traits and standing biomass

We expect the environmental variables to be more or less correlated (Fig. 1b). For instance, groundwater level and salinity ('Groundwater') should be strongly correlated with inundation frequency. By addressing these correlations (double-headed arrows), we also consider the

indirect effects of a variable of interest on other variables through correlations among the environmental variables.

Salinity and inundation frequency are considered the primary stress factors in salt marshes, among which salinity is regarded to as the ‘master factor’ (Rozema et al. 1985). Salinity affects biomass production and weight of the whole plant or roots and leaves of many salt marsh species (Lenssen et al. 1995; Egan & Ungar 2001), whereas for inundation frequency, different responses were reported, such as an increase (Lenssen et al. 1993) or decrease in biomass production or no response at all (Groenendijk 1985). Based on this background, we assume the ‘groundwater’ variables, level and salinity, will have negative effects on weight of leaves, stems, below-ground organs, MAOS, C:N ratio and SLA, and inundation frequency will have a negative effect on C:N ratio and MAOS.

Nutrient availability strongly affects primary productivity in salt marshes (Kiehl et al. 1997). We assume that nutrient availability (phosphorus, potassium and carbonate) will positively influence leaf, stem, diaspore, below-ground biomass and SLA (Cunningham et al. 1999; Poorter & de Jong 1999). Olff et al. (1997) has shown that nitrogen availability increases with silt content of salt marsh soils, which, in reverse, decreases with sand content. We therefore expect sand content to have a negative influence on SLA, leaf and stem biomass, and a positive influence on below-ground biomass. Following De Deyn et al. (2008), plant traits that determine carbon and nutrient conservation are, among others, high C:N ratio and long litter residence time, which is why we expect C:N ratio to positively explain dead biomass (Fig. 1c).

Above-ground biomass of the community (AGB) is the product of the biomass of leaves, stems and diaspores of the individual plants of the community. Therefore, we expect stem biomass and leaf biomass of the species to explain live AGB. Garnier et al. (2001) attributed high SLA to rapid production of biomass with high turnover rates; hence we expect a positive relationship between SLA and dead AGB. Finally, as dead AGB consists of formerly living biomass, we expect a positive relationship between live and dead AGB.

## Methods

### Study area

The study was conducted at three study areas along the mainland coastline of Lower Saxony, Germany, and on the island of Mellum (52°43′N, 8°08′E, 41 plots). On the mainland, fieldwork was carried out in three study areas: Leybucht (53°32′N, 7°07′E, 8 plots), Norderland (53°40′N, 7°19′E, 32 plots) and Jade Bight (53°26′N, 8°09′E, 32 plots). Mean annual temperature is about

9 °C and annual precipitation ranges from west to east from 770 to 830 mm yr<sup>-1</sup> (based on data from 1961 to 1990 for Norderney and Wilhelmshaven, Deutscher Wetterdienst 2009).

The foreland marshes of Lower Saxony often developed through land reclamation, extending over an area of 5430 ha (Pott 1995; Bakker et al. 2005), and consist predominantly of clayey silt, loamy sand and loamy silt. The island of Mellum originated from sand deposits transported by currents along the coast from The Netherlands and Northwest Germany (Pott 1995). It now belongs to the core zone of the Wadden Sea National Park of Lower Saxony. Whereas some soils consist of silty clay and clayey loam, the predominant substrate is sand on the island.

### Sampling design

The selection of sites was based on stratification of the salt marshes of Northwest Germany, in relation to human-modified versus natural conditions, silt versus sand and unused versus grazed or mown. Mellum was considered to be natural, with sandy soils, and unused land. The three mainland areas differed with respect to management. Within the study areas, the distribution of plots was based on random stratified sampling, with elevation as the stratification criterion.

Plots were sampled in summer 2007. At each of the 113 plots, we collected soil samples from each soil horizon. Soil sampling was restricted to a depth of 30 cm due to upwelling groundwater. Soil samples were air dried, sieved through a 2-mm sieve and analysed for sand content (Ad-Hoc-AG Boden 2005) and calcium carbonate (CaCO<sub>3</sub>, according to Scheibler in Schlichting et al. 1995). Soil available potassium and phosphorus were extracted with ammonium lactate-acetic acid at pH 3, following Egnér et al. (1960), and analysed using AAS (atomic absorption spectroscopy) and CFA (continuous flow analysis, Murphy & Riley 1962), respectively. Groundwater level and salinity were measured from May to September 2007. Looped drainage pipes allowing water inflow were buried vertically in the ground at each survey plot. Groundwater level was recorded bi-weekly at low tide in the drainage pipes. Data were revised by regression analysis to generate mean values including high tide events, see Statistical Analysis. Salinity of groundwater was recorded using a conductivity measurement device (‘WTWpH/Cond 340i/SET’ with measuring electrode ‘Tetracon 325’). Times series of both groundwater level and salinity were aggregated to their mean between May and September 2007.

Data loggers (‘diver’, ecoTech, Pegel-Datenlogger PDLA) were installed at 18 plots of different elevations to

**Table 1.** Environmental, trait and ecosystem property variables used for this study. Carbonate, potassium and phosphorus were aggregated by PCA analysis; sample scores were named Nutrients. The same was done for groundwater level and salinity; sample scores were named Groundwater. Diaspores, stems, leaves and belowground biomass values were log-transformed prior to analyses. Mean values and standard deviation (SD) for all variables on mainland and island plots are given in the last columns.

	Abbreviation	Unit	Transf.	Mainland		Island	
				Mean	SD	Mean	SD
Environmental variables							
Carbonate		t ha <sup>-1</sup>	None	207.15	66.57	18.81	22.64
Potassium	Nutrients	kg ha <sup>-1</sup>	None	1858.55	607.04	853.75	333.98
Phosphorus		kg ha <sup>-1</sup>	None	184.49	57.82	100.42	56.05
Mean level of groundwater	Groundwater	cm	None	-30.97	12.52	-21.41	16.44
Mean salinity of groundwater		PSU	None	22.47	4.75	24.87	3.36
Inundation frequency	Inundation	hour	None	172.82	273.97	242.39	297.94
Soil sand content	Sand	%	None	44.31	20.89	73.26	20.31
Trait variables							
Dry weight of							
Diaspore unit	Diaspore	g	log	1.79	1.20	0.58	0.44
Stems	Stem	g	log	7.78	8.69	1.99	1.79
Leaves	Leaves	g	log	1.27	0.62	1.88	1.61
Below-ground organs	Belowground	g	log	1.50	1.11	4.72	4.92
C:N ratio of whole plant	C:N ratio	None	None	37.98	7.56	35.54	5.51
Specific leaf area	SLA	mm <sup>2</sup> mg <sup>-1</sup>	None	12.06	2.79	9.40	2.52
Morphological adaptation to							
Osmotic stress	MAOS	1-Yes/0-No	None	0.37	0.33	0.25	0.38
Ecosystem property variables							
Above-ground biomass live	AGB live	g m <sup>-2</sup>	None	466.99	191.22	229.59	164.95
Above-ground biomass dead	AGB dead	g m <sup>-2</sup>	None	182.64	104.98	94.21	91.35

record inundation frequency. They were positioned at the bottom of the drainage pipes and recorded the salient water column on an hourly basis. An additional four data loggers recorded pressure of the surrounding air, which was used to calculate the relative pressure of water accumulating in the pipe. Almost all abiotic and trait variables differed strongly between mainland and island salt marshes (Table 1). Thus, mainland and island marshes were considered different habitats for this study.

Above-ground biomass was sampled in August, which is the time of peak biomass (De Leeuw et al. 1990). Samples were collected in an area of 0.5 m<sup>2</sup> on each survey site, and sorted into live and dead plant material. Samples were oven dried at 70 °C for 72 hr and weighed. Species composition and abundance was evaluated by frequency analysis using a 1 m × 1 m frame subdivided into 100 grids of 0.1 m × 0.1 m. Nomenclature followed Flora Europaea in the Syn-BioSys Species Checklist (2010).

Plant functional traits were sampled for 242 individuals, i.e. approximately 10 individuals per species. These individuals were selected from different plots covering the total range of species occurrences within the environmental space. For measurement of biomass allocation, plants were collected at the peak of their generative stage, i.e. when seeds were ripe but not yet shed. These data

were used for calculation of the community-weighted means of stem, leaf, diaspore and below-ground biomass (see below). In addition, we collected 112 individuals from 14 mainland species at the peak of their vegetative stage, i.e. before flowering. Plants were dug out, roots and rhizomes were cleaned of soil material by rinsing off the soil substrate, and roots of different individuals were carefully separated using tweezers. Plant material was subsequently oven dried at 70 °C for 72 hr. Leaves, stems, diaspores and below-ground organs (roots and rhizomes) were weighed after drying. C:N content was measured after grinding the plant material in a planetary mill for 2 to 10 min at 300 to 400 revolutions (pulverisette 7, Fritsch). Each sample was then dried at 105 °C for 4 to 5 hr. Two to three milligrams of material were placed in tin tubes (0.1 mg precision balance CP 225 D, Sartorius; tin capsules for solids, Sántis Analytical) and analysed using a CHNS analyser Flash EA (Thermo Electron Corp.) following Allen (1989).

SLA (specific leaf area) was calculated as the ratio of leaf area to leaf dry mass (mm<sup>2</sup> mg<sup>-1</sup>) following Knevel et al. (2005). The trait morphological adaptation to osmotic stress (MAOS) was evaluated using a literature search, with subsequent assignment of either existence (1) or non-existence (0) of this attribute to a species.

## Statistical analysis

### *Groundwater level*

At all plots, groundwater level was recorded bi-weekly during low tide and thus contained no information about variation due to tidal elevation. Therefore, a regression was conducted with paired values of the hourly data from the data loggers at a subset of 18 plots and the bi-weekly data of groundwater level measured at these plots. Mean values of both groundwater and data loggers over time were generated for each of these plots and used for linear regression analysis. The regression function was used to adjust values of mean groundwater level of all other plots to include information about tidal effects.

### *PCA of environmental parameters*

There were strong correlations between carbonate, potassium and phosphorus, as well as between groundwater level and groundwater salinity. Therefore, to reduce the number of variables in the model, and using principal component analysis (PCA), carbonate, plant available potassium and phosphorus were aggregated to their first principal component called ‘nutrients’. Likewise, groundwater level and salinity were aggregated to ‘groundwater’. Analysis was carried out using the computer software R (version 2.8.1, The R Foundation for Statistical Computing 2008); variables were standardized as they were measured at different scales.

### *Bivariate scaling relationships at the individual level*

Standardized major axis regression (SMA, Warton et al. 2006) was used to describe bivariate relationships between traits at the individual level. Trait variables are biologically interdependent and have similar variation associated with them due to measurement error. In this case, ordinary least squares regression is considered inappropriate (Niklas 1994), because it keeps the  $x$ -values fixed, and finds the line that minimizes the squared errors in the  $y$ -values, whereas SMA tries to minimize both the  $x$ - and  $y$ -errors. SMA regression was done with the software PAST (version 1.94b, Hammer et al. 2001).

### *Community-weighted mean*

According to Grime (1998), the extent to which a plant species influences ecosystem function is likely to be derivable from its contribution to total plant biomass, which is indicated by its abundance. To predict AGB from traits, we therefore used community-weighted means (Garnier et al. 2007). The mean value of each trait of every species was weighted by the abundance of the species on each plot and averaged by the total abundance of every species on each plot. A list of all variables used in the path analysis is given in Table 1.

### *Path analysis and structural equation modelling (SEM)*

Path analysis was used to quantify the relationships among variables of our a priori model, i.e. the relations between abiotic conditions, plant functional traits and ecosystem functioning (standing biomass). The effect–response framework of Lavorel & Garnier (2002) is a uni-directional framework (from abiotic conditions to ecosystem properties via functional traits) thus it was decided not to consider reciprocal paths (e.g. paths from ecosystem property to functional traits) in the analysis. We used path analysis to partition the correlations among observed variables and to measure both direct and indirect effects on response variables (Kingsolver & Schemske 1991; McCune & Grace 2002). SEM was used to test overall model fit (Grace & Pugsek 1998).

The hypothetical model (Fig. 1) generates an expected covariance structure, which is compared to the actual covariance matrix. Standardized coefficients describe the strength of the relationship. Relationships between variables are either uni-directionally causal (indicated by a straight, single-headed arrow) or unanalysed correlations (indicated by a curved, double-headed arrow). Indirect pathways between variables involve intermediary variables. Direct pathways between two variables are the value of path coefficients, which are the standardized partial regression coefficients (Grace 2006).

SEM allows testing of the hypothesis that the measured covariance structure adequately describes the expected covariance structure. This is done by means of maximum likelihood estimates, which generate a test statistic that is distributed approximately as  $\chi^2$  (Mitchell 1992). Good fit of the hypothesized model to the data will result in low  $\chi^2/df$  and non-significant  $P$ -value. Goodness-of-fit-index (GFI) is a measure of the relative amount of variance and covariance that the model allows. Another index of fit, the root mean square error of approximation (RMSEA, Browne & Cudeck 1993), assesses closeness to fit. Good models have  $RMSEA < 0.05$ ,  $P > 0.05$ ,  $\chi^2/df$  values  $< 2$ , and  $GFI > 0.9$  (Backhaus et al. 2003). Path analysis and SEM was done using Amos 16.0.1 (Amos Development Corporation Spring House, PA, US).

## Results

### Environmental and ecosystem property variables

A comparison between mainland and island revealed significant differences between all environmental parameters at  $P > 0.05$  (ANOVA, Table 1). Soils of mainland marshes were richer in nutrients and contained less sand than those of island marshes. AGB was higher in mainland marshes than in island marshes. High standard deviations resulted partly from the orientation of the plots along the elevation gradient; for example, plots in the

pioneer zone were inundated for about 100 to 600 hr during the period 13 May to 17 September, whereas plots in the upper marsh were only inundated for about 3 to 100 hr. As there were more plots in the lower salt marshes of the island than of the mainland, mean inundation frequency was higher in island marshes.

### Trait variables

Sixteen different species were sampled, which represented about 90% of the regional salt marsh species pool. Four were grasses (*Spartina anglica*, *Puccinellia maritima*, *Festuca rubra* and *Elymus athericus*) and two were perennial sub-shrubs that develop woody stems (*Artemisia maritima* and *Atriplex portulacoides*). The remaining species were annual and perennial herbs.

Trait values for the community on the mainland and the island differed significantly for diaspore, stem and below-ground weight, as well as for MAOS and SLA at  $P > 0.05$  (Table 1). Only C:N ratio and leaf biomass showed no significant differences. High standard deviations resulted from different trait expressions along the elevation gradient, e.g. *Aster tripolium* of the lower marsh produced higher stem biomass (mean 41.9 g for mainland) than *Elymus athericus* from the higher marsh (mean 0.32 g for mainland).

### PCA of environmental parameters

Correlation coefficients for nutrients were high (potassium – carbonate: 0.67, potassium – phosphorus: 0.53, and carbonate – phosphorus: 0.65) and mean groundwater level and salinity also showed a high correlation (0.64). All correlation coefficients were significant at  $P > 0.01$  level.

Explained variance for nutrients (i.e. the first principal component of potassium, phosphorus and carbonate) and groundwater (the first principal component of groundwater level and salinity) are high for the first axis (0.74 and 0.82) and accordingly lower for the second (0.16 and 0.18, Fig. 2). PCA sample scores for nutrients and groundwater were used for further path analysis.

### Biomass partitioning at the individual level

At the individual level, log-log relationships between pairs of biomass of organs were close to isometry (Table 2). Only the relationships between below-ground biomass (roots and rhizomes) and leaf and diaspore mass were significantly different from isometry, for the vegetative and generative stage. For the generative stage,  $r^2$  values were lower than for the vegetative stage. We assumed that the variation in these bivariate relationships, as expressed by the  $r^2$ , reflect environmental constraints. Therefore, we decided to include biomass of all plant organs in the path model even though they were correlated.

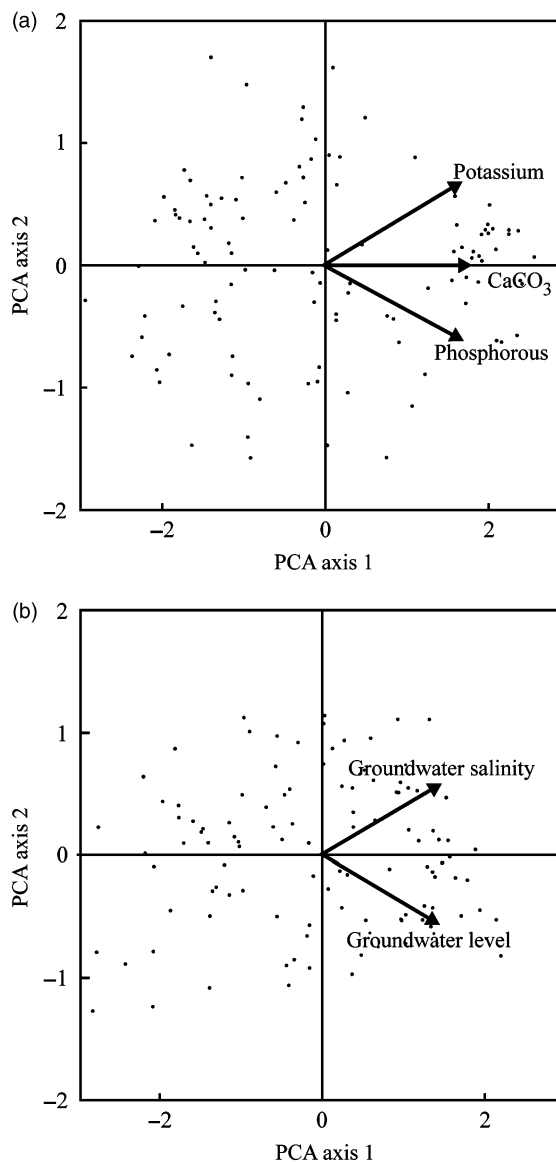


Fig. 2. PCA of environmental parameters using potassium, carbonate and phosphorus (a) and groundwater salinity and groundwater level (b).

### Path analysis and structural equation modelling (SEM)

Mardia's coefficient (Mardia 1970; Mardia 1974) suggested a non-normal distribution of the data of the initial model. To obtain multivariate normality, nine sample points were deleted, using Mahalanobis distance as indicator. Several paths were added to improve model fit (nutrients → MAOS; SLA → diaspore; stem → dead AGB; C:N ratio → diaspore; below-ground → diaspore, MAOS, live AGB, dead AGB).

Several direct paths of the initial model proved non-significant, however most of them showed significant total effects (see Table 3), these were: groundwater → below-

**Table 2.** Statistics of pair-wise reduced major axis regression analyses (RMA) of standing leaf, stem, below-ground and diaspore biomass ( $M_L$ ,  $M_S$ ,  $M_R$  and  $M_D$ , respectively).  $M_R$  comprises root and rhizome masses. The slope of the regression is denoted by  $\alpha_{SMA}$  and the intercept by  $\beta_{SMA}$ .  $P(\alpha = 1)$  shows the probability that slope  $\alpha$  of the regression is isometric. In all cases,  $P < 0.0001$ .

$Y_2$ versus $Y_1$	$\alpha_{SMA}$	SE	$\beta_{SMA}$	SE	95%CI	$r^2$	$n$	$P(\alpha = 1)$
Peak vegetative stage								
$M_L$ versus $M_S$	0.94	0.05	-0.079	0.040	0.85–1.03	0.73	112	0.217
$M_S$ versus $M_R$	0.96	0.06	0.263	0.061	0.85–1.08	0.57	112	0.528
$M_L$ versus $M_R$	0.90	0.04	0.167	0.046	0.80–0.99	0.72	112	0.042
Peak generative stage								
$M_L$ versus $M_S$	0.98	0.05	-0.393	0.042	0.89–1.09	0.36	242	0.742
$M_S$ versus $M_R$	0.91	0.05	0.142	0.040	0.82–1.01	0.35	242	0.060
$M_L$ versus $M_R$	0.89	0.04	-0.252	0.038	0.83–0.95	0.52	242	0.009
$M_D$ versus $M_R$	0.86	0.04	-0.277	0.037	0.79–0.94	0.51	242	0.001
$M_D$ versus $M_L$	0.97	0.05	-0.032	0.047	0.87–1.07	0.37	242	0.570
$M_D$ versus $M_S$	0.95	0.03	-0.413	0.025	0.90–1.01	0.75	242	0.147

ground; inundation → MAOS; nutrients → leaf; leaf → diaspore; below-ground → stem.

The changes made to the initial model led to a stable model that only included essential pathways, which were all significant at  $P < 0.05$ . This model showed good consistency with the data ( $\chi^2/df = 1.022$ ,  $P = 0.431$ , GFI = 0.947, RMSEA = 0.015) and explained 45% of the variation in live AGB and 48% in dead AGB.

The SEM revealed both direct and indirect effects between environmental conditions, traits and above-ground biomass. Direct effects are visualized in Fig. 3a–c; indirect effects occur if two variables are connected through paths to and from a third variable. Total effects are calculated from the combined direct and indirect effects (Table 3). Here, we concentrate on relevant total effects, i.e. those  $> 0.3$  in Table 3.

### Trait–trait linkages

Most of the relationships of the final model confirmed our expectations of the initial model. Some direct relationships of the initial model proved non-significant; however, total effects between these traits justified the initial expectations (e.g. direct relationship between below-ground and stem was not significant, total effect was +0.48, Table 3). In contrast, we expected SLA and stem mass, as well as C:N ratio and leaf mass to be positively related, which proved correct for the direct effect (+0.33 and +0.61) but not for the total effect (-0.19 and -0.18). These results highlight the influence of indirect causes on plant traits and the importance of considering the total effects between two variables, rather than solely concentrate on direct effects.

Four paths were added to the initial model, of which the strongest were between below-ground and diaspore mass (total effect 0.68), and C:N ratio and below-ground mass (-0.35). According to the first path, reproductive output increases with below-ground biomass. In contrast, plants with high C:N ratio have low below-ground mass,

which then means that those plants do not invest heavily in diaspore production.

The strongest allometric relationships confirming the initial expectations were between below-ground organs and leaf mass, stem mass and SLA and between stem and diaspore mass and leaf and stem mass (total effects Table 3). C:N ratio showed a positive relationship with MAOS, and the strongest trade-off was between MAOS and leaf mass, i.e. in the upper marsh, tissue of plants is rich in carbon and allocation towards leaves is low. The C:N ratio showed a strong trade-off with SLA and below-ground mass, which means that, considering the relationships already mentioned, upper marsh species have low SLA values and low allocation to below-ground organs.

### Response traits

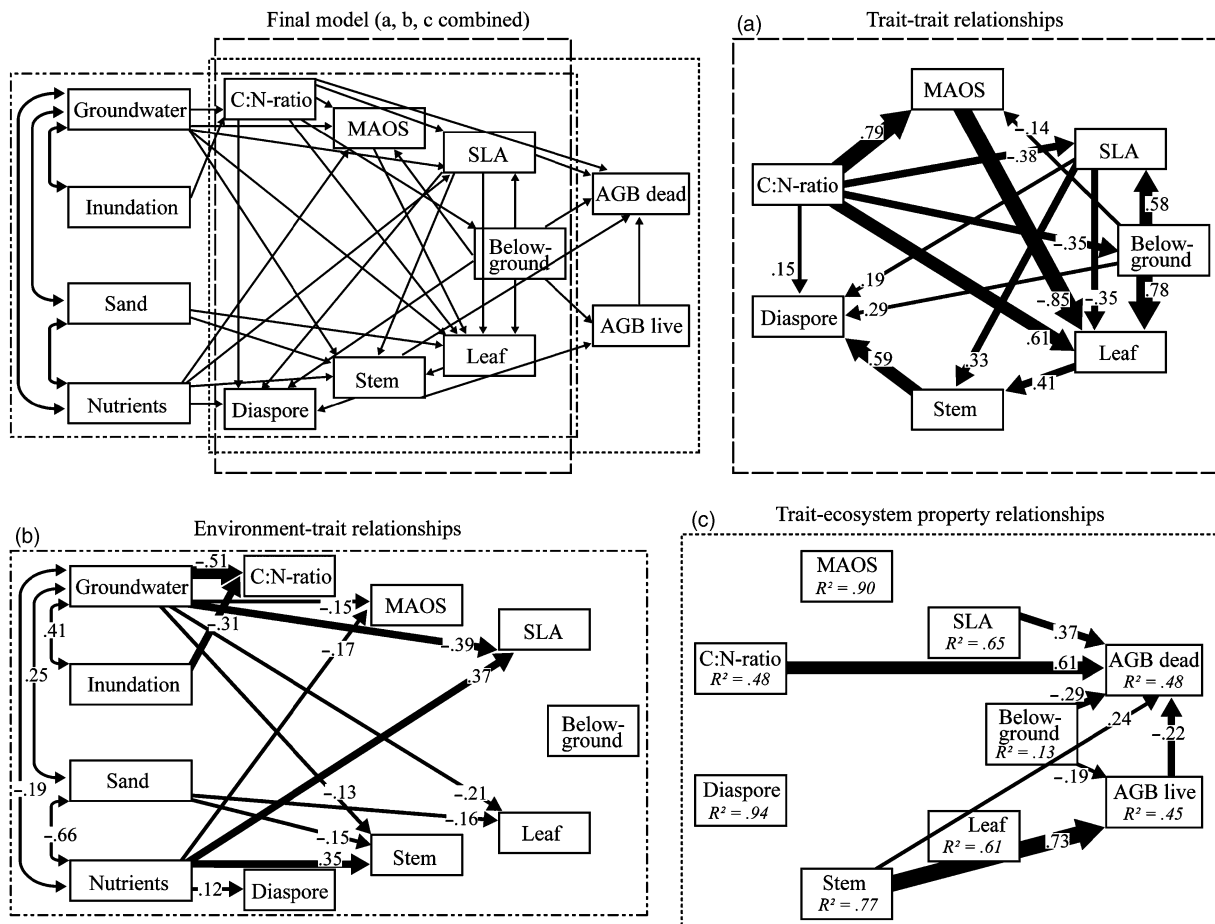
Sand was negatively correlated to nutrients and positively to groundwater, whereas groundwater was positively correlated to inundation frequency (Fig. 3b). This indicates that the lower parts of the salt marsh, with high inundation frequency, are less rich in nutrients than the upper parts.

The traits responding most strongly were SLA, stem and diaspore mass, with positive responses to nutrients (total effects Table 3, direct effects Fig. 3b), C:N ratio and MAOS, and with strong negative responses to groundwater and inundation frequency (the latter with response from C:N ratio only). There were no relevant responses to sand content.

### Effect traits

Stem biomass, together with leaf mass, had the strongest positive effects on standing live above-ground biomass, which confirmed our expectations. However, there was no direct relationship between leaf mass and AGB live (Fig. 3c). Leaves exerted an influence on standing live biomass through an allometric relationship with stem mass. C:N ratio and SLA had the strongest negative total





**Fig. 3.** Final model (upper left) derived from initial model in Fig. 1. From the final model, relationships between traits (a, upper right), environment–trait relationships (b, bottom left) and trait–ecosystem property relationships (c, bottom right) were magnified for a better overview. Names and abbreviations of observed variables follow Table 1. Path coefficients between variables are standardized partial regression coefficients of direct effects, for total and indirect effects, see Table 3. Arrow widths are proportional to the standardized path coefficient. Variances explained by the model ( $R^2$ ) are given under the variable names (c). Residual errors were omitted. All pathways are significant at  $P < 0.05$ .

effects on AGB dead. Hence, stem biomass, SLA and C:N ratio were both response and effect traits.

#### Indirect effects of environmental variables on standing biomass

Through their effects on traits, groundwater had a negative indirect effect on above-ground dead biomass, and nutrients had a positive but weak indirect effect on above-ground live biomass.

#### Discussion

Lavorel & Garnier (2002) referred to linkages between environment, biodiversity and ecosystem properties via response and effect traits as the ‘holy grail’ of functional ecology. Path analysis and structural equation modelling allowed us to quantify these linkages for a single ecosystem property: above-ground biomass of salt marshes. In particular, we were able to demonstrate that the allo-

metric biomass allocation pattern at the species level translates into the community level. Through the allometric relationship among stem, leaf, below-ground and diaspore mass, a single response and effect trait, stem biomass, drove the relationship between soil nutrients and above-ground live community biomass. On the other hand, whole-plant C:N ratio and SLA were the main response and effect traits driving the relationship between above-ground dead community biomass and groundwater level, salinity and nutrients.

#### Scaling relationships in plant biomass allocation at the species level

To understand whether the SEM relationships among plant organ biomasses at the community level are consistent with those at the species level, we investigated interspecific allometric scaling relationships at the vegetative and generative stages. Most of these relationships

**Table 3.** Standardized total, direct and indirect effects of factors that influence above-ground biomass production. Path coefficients between variables are standardized partial regression coefficients. Total effects were calculated by adding direct and indirect effects. All effects were significant at  $P < 0.05$ .

	Groundwater			Inundation			Sand			MAOS			Nutrients			AGB live		
	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.
C:N ratio	-0.51	-0.51		-0.31	-0.31													
Below-ground	0.18		0.18	0.11		0.11												
SLA	-0.09	-0.39	0.29	0.18		0.18							0.37	0.37				
MAOS	-0.58	-0.15	-0.43	-0.24		-0.24							-0.17	-0.17				
Leaf	0.15	-0.21	0.36	0.06		0.06	-0.16						0.01		0.01			
Stem	-0.11	-0.13	0.03	0.08		0.08	-0.22	-0.15					0.48	0.35	0.13			
Diaspore	-0.11		-0.11	0.07		0.07	-0.13						0.48	0.12	0.36			
AGB live	-0.11		-0.11	0.04		0.04	-0.15						0.35		0.35			
AGB dead	-0.39		-0.39	-0.14		-0.14	-0.02						0.17		0.17	-0.22		-0.22
C:N ratio																		
Below-ground																		
Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.
C:N ratio	-0.35	-0.35																
Below-ground	-0.58	-0.38	-0.20	0.58	0.58													
SLA	0.84	0.79	0.05	-0.14	-0.14													
MAOS	-0.18	0.61	-0.79	0.70	0.78	-0.08	-0.35	-0.35	-0.85	-0.85								
Leaf	-0.26		-0.26	0.48		0.48	-0.19						-0.35	0.41	0.41			
Stem	-0.22	0.15	-0.37	0.68	0.29	0.39	-0.30	-0.11	-0.21	-0.21			-0.21	0.24	0.24	0.24	0.59	0.59
Diaspore	-0.12		-0.12	0.16	-0.19	0.35	-0.14	-0.14	-0.25	-0.25			-0.25	0.29	0.29	0.29	0.73	0.73
AGB live	-0.45	0.61	-0.15	-0.29	0.29	0.29	-0.38	-0.01	-0.03	-0.03			-0.03	0.03	0.03	0.08	0.24	0.24
AGB dead																		
Stem																		
Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.	Dir.	Ind.	Tot.

were close to isometry, whereas, according to allometric theory, they should scale to the three-quarter power, except for the relationship among root and stem mass, which should be isometric (Enquist & Niklas 2002; McCarthy & Enquist 2007). However, the three-quarter rule was empirically validated mainly for tree species, whereas deviations from this rule towards isometry were found for juvenile and herbaceous plants (Enquist & Niklas 2002; Niklas 2006). In our salt marsh species set, allometric relationships close to isometry were present at the vegetative and generative stages, and among vegetative and reproductive organs. In a recent review of allometric relationships among vegetative and reproductive biomass of herbaceous species, Weiner et al. (2009) found linear relationships close to isometry for the majority of the 76 species investigated. The scaling relationships at the species level suggest a common allometric strategy across all salt marsh species (Müller et al. 2000), implying that an increase in biomass of one organ is accompanied by a proportional increase in biomass of all other organs. However, the variation in the data indicated species-specific deviations from this general scaling rule, which may have strong effects at the community level when such species become dominant. Environmentally constrained species sorting may thus result in ecosystem properties that cannot be predicted from general scaling rules derived from the whole species pool (Suding et al. 2008).

#### Relationships among plant traits at the community level

At the community level, i.e. when the mean organ biomass of all species present in a community is weighted by their abundance, the modelled relationships among stem, leaf, diaspore, root and rhizome mass were also allometric (Table 3). Corresponding to our hypothesized model, we found strong positive total effects of below-ground biomass on leaf, stem and reproductive biomass, as well as of stem biomass on diaspore biomass. Total effects of leaf biomass on stem and diaspore mass were less pronounced. The path analysis and structural equation model explores these relationships in a multivariate rather than a bivariate way, which has rarely been undertaken (Shipley 2004).

Our expectation that leaf biomass would scale strongly negative with MAOS as a salt stress regulation mechanism was also confirmed. MAOS attributes to species of the upper salt marsh, which is a late successional stage in this habitat. Leaf mass proved a good predictor for partitioning the salt regulation mechanisms between the upper and lower salt marsh.

#### Trait responses to environmental conditions

Our final model illustrates that groundwater level, salinity and nutrient availability were the driving factors

influencing functional traits, while inundation frequency and sand content of the soil played minor roles.

Contrary to the hypothesized model, there were no direct links between nutrients and leaf or below-ground biomass. However, nutrients had a direct effect on SLA and on biomass allocation to stems. In addition, sand content, being strongly negatively correlated to nutrients in this study and considered as a proxy for low nitrogen availability in salt marshes (Olf et al. 1997), had a negative total effect on stems. With increasing nutrients, plants invest more into supporting stem tissue, thus seizing the favourable conditions to produce more vegetative tissue and to acquire dominance by occupying vertical space and better competing for light (Tilman 1988; Westoby et al. 2002). In a greenhouse experiment, Kuijper et al. (2005) showed that the above-ground relative yield of *Elymus athericus*, a dominant plant of the high salt marsh, increased with nitrogen availability at the expense of *Festuca rubra*. *Elymus* also invested relatively more biomass in stem and root tissue and had larger shoot length than *Festuca*. This experiment confirms that changes in nutrient supply affect stem biomass allocation, which results in changes in species abundances. Similar results were found in transplant experiments on Dutch salt marshes, where increasing soil nutrient levels indicated by decreasing sand and increasing clay content favoured species with comparably high investment in stem tissue, while rosette species decreased in abundance (Dormann et al. 2000).

Through the allometric relationship between stems and other plant organs, nutrients also indirectly favoured diaspore production (Niklas & Enquist 2003), thus increasing the chance of plants to recruit (Moles & Westoby 2006) and to explore new habitats via their propagules.

Groundwater had a negative influence on C:N ratio and MAOS, i.e. plants growing at high groundwater levels and salinities tend to stock relatively less carbon than nitrogen compared to those subjected to more balanced water conditions. This is in contrast to other terrestrial conditions, where plants growing in more stressful habitats are often characterized by higher C:N ratios (De Deyn et al. 2008). However, many plants capable of withstanding salt stress and frequent inundation synthesize nitrogen-requiring osmoprotectants, such as proline, glycine betaine and other quaternary ammonium compounds, to counter osmotic stress (Steward et al. 1979; Cavalieri 1983; Tarczynski et al. 1993). These osmotic solutes often make up a large part of the plant nitrogen budget (Roze-ma et al. 1985).

The relationship between inundation frequency and groundwater level and salinity was not as straightforward as expected, because the correlation between these two parameters was only moderate (0.41). We

found that less frequently inundated higher salt marshes also had high groundwater salinities. In these cases, high salinity may arise from summer rainfall deficits (precipitation minus evapotranspiration), which cause decreased water content of the sediment and hypersaline conditions of the soil substrate (Leeuw et al. 1990). This may explain why C:N ratio responded more strongly to groundwater level and salinity than to inundation frequency.

### Keystone response and effect traits

Our model detected four traits explaining the community standing biomass. These are C:N ratio and SLA explaining dead biomass, on the one hand, and leaf and stem mass, which affect living biomass, on the other hand. Of these traits, C:N ratio, SLA and stem mass were both effect and response traits.

Live biomass is explained by stem biomass because, based on the isometric allometry of all above-ground organs including leaf mass, an increase in stem biomass leads to a concurrent biomass increase in all other organs. On the other hand, dead biomass is determined by the plant C:N ratio, with increasing carbon stocks leading to a higher dead fraction of the total biomass. We assume that the mechanism behind this link is an accumulation of litter due to reduced decomposition in the upper marsh. This assumption was confirmed by decomposition studies performed for a subset of the plots, showing that the litter of plants with a high C:N ratio, particularly *Elymus athericus*, decomposed more slowly than plant litter with a lower C:N ratio (data not shown). Species from early successional stages tend to decompose more rapidly than those from more advanced stages (Garnier et al. 2004; Kazakou et al. 2006). In salt marshes, *Elymus athericus* forms the successional climax of the upper marsh (Bakker et al. 2003).

De Deyn et al. (2008) showed that when soil resources are reduced, e.g. water, traits that drive carbon and nutrient conservation dominate, such as high C:N ratio and longer litter residence time (Aerts & Chapin 2000). Our study showed that a high groundwater level and salinity negatively influence C:N ratio, i.e. high groundwater leads to low C:N ratios, as the demand for nitrogen for osmoregulation increases at high levels of groundwater. We assume that plants of the upper marsh demand nitrogen for osmoregulation to a lesser extent than those of the lower marsh and the pioneer zone. Thus, in the upper marsh, C:N ratio is higher but litter decomposition is also reduced, which leads to accumulation of dead biomass.

The second trait to explain dead biomass is SLA, although the effect on AGB dead was not as pronounced

as the effect of C:N ratio. SLA is higher in the lower marsh, which is expressed by the strong negative relationship to C:N ratio. Fast-growing species from nutrient-rich sites have high SLA together with high tissue nutrient content and short-lived leaves (Lavorel et al. 2007). We assume that species of the lower marsh show high production of biomass, but as the turnover rate of this biomass is high, the proportion of dead biomass increases. Dead plant material in the lower marsh can be washed away more easily by incoming water or spring tides than in the upper marsh, so that an accumulation of dead biomass in the upper parts of a salt marsh relative to the lower parts seems obvious. However, Bakker et al. (1993) estimated decomposition rates in Dutch salt marshes and found that tissue composition (especially lignin content) was the most important factor determining rates of decomposition, and environment heterogeneity (water, silt and salt content of the soil) was only the second most important factor. This supports our assumption that the weaker effect of SLA on dead biomass in comparison to the C:N ratio is a combination of high production of biomass of the lower marsh species, on the one hand, and higher losses due to decomposition on the other.

### Conclusions

The relationship between standing biomass and environmental conditions has often been addressed as a direct dependency. Here, we emphasize the indirect link, i.e. plant traits respond to the environment and simultaneously affect above-ground standing biomass. Keystone response and effect traits were (i) stem biomass responding to available nutrients and affecting standing live biomass, as well as (ii) plant C:N ratio and SLA responding to groundwater (level and salinity) and nutrients and affecting standing dead biomass. These findings of response and effect traits did not come as a surprise. It is evident, yet rarely shown, that stem biomass drives standing live biomass via an underlying isometric allocation of biomass to all other plant organs, including leaves, conforming to allometric theory. Nitrogen content and SLA are closely related to maximum rate of photosynthesis across plants worldwide, and are thus important and rather simple measurable parameters of plant growth (Wright et al. 2004). The relevance of whole plant C:N ratio in determining dead biomass has been confirmed for many different ecosystems. SLA was not merely a counterpart of C:N ratio, because the former was more determined by soil resources and the latter more by salt and aeration. Generally, these traits can be considered keystone markers of species functioning in determining the relationship between environment and ecosystem properties. The identification of a relatively small set of key

traits translating the relationship between environment and above-ground biomass through the species and community level is a relevant step in the theoretical understanding, modelling and managing of biodiversity for ecosystem properties. For instance, basing the classification of plant functional types on such traits rather than on coarse combinations of life form and phenology may improve predictions of dynamic global vegetation models (Harrison et al. 2010). Regarding management as well as biomass production, these traits are associated with multiple ecosystem services, such as carbon sequestration (De Deyn et al. 2008) or fodder quality (Pontes et al. 2007). The most relevant salt marsh-specific result was the inverse relationship between C:N ratio and environmental stress, as compared to most other terrestrial ecosystems. Lower C:N ratio in plants with increasing salt stress is apparently due to higher nitrogen investment to synthesize osmoprotectants under salt stress.

### Acknowledgements

We thank the administration of the National Park 'Niedersächsisches Wattenmeer' and 'Mellumrat e.V.' for their support during fieldwork. Many thanks to H. Timmermann and J. Spalke for contributing their data, S. Andratschke for field assistance, G. Schweiffarth and M. Heckroth for supporting our work at Mellum, and E. Garnier for valuable comments on previous versions of the manuscript. This study was conducted as part of the TREIBSEL project and was financially supported by the 'II. Oldenburgischer Deichband' and the 'Wasserverbandstag e.V.' (NWS 10/05).

### References

- Ad-Hoc-AG Boden 2005. *Bodenkundliche Kartieranleitung*. E. Schweitzerbart'sche Verlagsbuchhandlung, Hannover, DE.
- Aerts, R. & Chapin, F.S.I. 2000. The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. *Advances of Ecological Research* 30: 1–67.
- Allen, S.E. 1989. *Chemical analysis of ecological materials*. Blackwell Scientific, Oxford, UK.
- Backhaus, K., Erichson, B., Plinke, W. & Weiber, R. 2003. *Multivariate analyse methoden*. Springer-Verlag, Berlin, DE.
- Bakker, J.P., De Leeuw, J., Dijkema, K.S., Leendertse, P.C., Prins, H.H.T. & Rozema, J. 1993. Salt marshes along the coast of The Netherlands. *Hydrobiologia* 265: 73–95.
- Bakker, J.P., Bos, D., Stahl, J., De Vries, Y. & Jensen, A. 2003. Biodiversität und Landnutzung in Salzwiesen. *Nova Acta Leopoldina NF 87* 328: 163–194.
- Bakker, J.P., Bunje, J., Dijkema, K.S., Frikk, J., Hecker, N., Kers, B., Körber, P., Kohlus, J. & Stock, M. 2005. Salt marshes. In: Essink, K., Dettman, C., Farke, H., Laursen, K., Lüerßen, G., Marencic, H. & Wiersinga, W. (eds.) *Wadden Sea quality status report 2004, Wadden Sea ecosystem No. 19 – 2005*. Common Wadden Sea Secretariat (CWSS), Wilhelmshaven, DE.
- Bazzaz, F.A. 1997. Allocation of resources in plants: state of the science and critical questions. In: Bazzaz, F.A. & Grace, J. (eds.) *Plant resource allocation*. pp. 1–37. Academic Press, San Diego, CA, US.
- Blew, J., Günther, K., Laursen, K., van Roomen, M., Südbeck, P., Eskildsen, K., Potel, P. & Rösner, H.-U. 2005. Overview on trend and numbers of migratory waterbirds in the Wadden Sea 1980–2000. In: Blew, J. & Südbeck, P. (eds.) *Migratory water birds in the Wadden Sea 1980–2000*. Common Wadden Sea Secretariat (CWSS), Wilhelmshaven, DE.
- Browne, M. & Cudeck, R. 1993. Alternative ways of assessing equation model fit. In: Boollen, K.A. & Long, J.S. (eds.) *Testing structural equation models*. pp. 136–162. Sage Publications, Newbury Park, CA, US.
- Cavaliere, A.J. 1983. Proline and glycine betaine accumulation by *Spartina alterniflora* Loisel. in response to NaCl and nitrogen in a controlled environment. *Oecologia* 57: 20–24.
- Cunningham, S., Summerhayes, B. & Westoby, M. 1999. Evolutionary divergences in leaf structure and chemistry, comparing rainfall and soil nutrient gradients. *Ecological Monographs* 69: 569–588.
- de Bello, F., Lavorel, S., Diaz, S., Harrington, R., Cornelissen, J.H.C., Bardgett, R.D., Berg, M.P., Cipriotti, P., Feld, C.K., Hering, D., da Silva, P.M., Potts, S.G., Sandin, L., Sousa, J.P., Storkey, J., Wardle, D.A. & Harrison, P.A. 2010. Towards an assessment of multiple ecosystem processes and services via functional traits. *Biodiversity and Conservation* 19: 2873–2893.
- De Deyn, G.B., Cornelissen, J.H.C. & Bardgett, R.D. 2008. Plant functional traits and soil carbon sequestration in contrasting biomes. *Ecology Letters* 11: 516–531.
- De Leeuw, J., Olf, H. & Bakker, J.P. 1990. Year-to-year variation in peak above-ground biomass of six salt-marsh angiosperm communities as related to rainfall deficit and inundation frequency. *Aquatic Botany* 36: 139–151.
- Deutscher Wetterdienst 2009. *Mean climate values for the period 1961 to 1990*. Available at [http://www.dwd.de/bvbw/appmanager/bvbw/dwdwwwDesktop?\\_nfpb=true&\\_pageLabel=\\_dwdwww\\_menu2\\_leistungen\\_a-z\\_freimetinfos&T115202758871200642573928gsbDocumentPath=Navigation%2FOeffentlichkeit%2FKlima\\_Umwelt%2FKlimadatenzentren%2FNKZ%2FKdaten\\_akt%2Fausgabe\\_mittelwerte\\_node.html%3F\\_nnn%3Dtrue](http://www.dwd.de/bvbw/appmanager/bvbw/dwdwwwDesktop?_nfpb=true&_pageLabel=_dwdwww_menu2_leistungen_a-z_freimetinfos&T115202758871200642573928gsbDocumentPath=Navigation%2FOeffentlichkeit%2FKlima_Umwelt%2FKlimadatenzentren%2FNKZ%2FKdaten_akt%2Fausgabe_mittelwerte_node.html%3F_nnn%3Dtrue) (accessed 6 April 2009).
- Díaz, S. & Cabido, M. 2001. Vive la difference: plant functional diversity matters to ecosystem processes. *Trends in Ecology & Evolution* 16: 646–655.
- Dormann, C.F., Van der Wal, R. & Bakker, J.P. 2000. Competition and herbivory during salt marsh succession: the importance of forb growth strategy. *Journal of Ecology* 88: 571–583.
- Egan, T.P. & Ungar, I.A. 2001. Competition between *Salicornia europaea* and *Atriplex prostrata* (Chenopodiaceae) along an experimental salinity gradient. *Wetlands Ecology and Management* 9: 457–461.

- Egnér, H., Riehm, H. & Domingo, W.R. 1960. Untersuchungen über die Bodenanalyse als Grundlage für die Beurteilung des Nährstoffzustandes des Bodens II. Chemische Extraktionsmethoden zur Phosphor- und Kaliumbestimmung. *Kungl. Lantbrukshögskolans Annaler* 26: 199–215.
- Enquist, B.J. & Niklas, K.J. 2002. Global allocation rules for patterns of biomass partitioning in seed plants. *Science* 295: 1517–1520.
- Garnier, E., Shipley, B., Roumet, C. & Laurent, G. 2001. A standardized protocol for the determination of specific leaf area and leaf dry matter content. *Functional Ecology* 15: 688–695.
- Garnier, E., Cortez, J., Billès, G., Navas, M.-L., Roumet, C., Debussche, M., Laurent, G., Blanchard, A., Aubry, D., Bellmann, A., Neill, C. & Toussaint, J.-P. 2004. Plant functional markers capture ecosystem properties during secondary succession. *Ecology* 85: 2630–2637.
- Garnier, E., Lavorel, S., Ansquer, P., Castro, H., Cruz, P., Dolezal, J., Eriksson, O., Fortunel, C., Freitas, H., Golodets, C., Grugulis, K., Jouany, C., Kazakou, E., Kigel, J., Kleyer, M., Lehsten, V., Lepš, J., Meier, T., Pakeman, R., Papadimitriou, M., Papanastasis, V.P., Quested, H., Qué-tier, F., Robson, M., Roumet, C., Rusch, G., Skarpe, C., Sternberg, M., Theau, J.-P., Thébault, A., Vile, D. & Zarovali, M. 2007. Assessing the effects of land-use change on plant traits, communities and ecosystem functioning in grasslands: a standardized methodology and lessons from an application to 11 European sites. *Annals of Botany* 99: 967–985.
- Gitay, H. & Noble, I.R. 1997. What are functional types and how should we seek them? In: Smith, T.M., Shugart, H.H. & Woodward, F.I. (eds.) *Plant functional types: their relevance to ecosystem properties*. pp. 3–19. Cambridge University Press, Cambridge, UK.
- Grace, J.B. 2006. *Structural equation modeling and natural systems*. Cambridge University Press, Cambridge, UK.
- Grace, J.B. & Pugsek, B.H. 1997. A structural equation model of plant species richness and its application to a coastal wetland. *The American Naturalist* 149: 436–460.
- Grace, J.B. & Pugsek, B.H. 1998. On the use of path analysis and related procedures for the investigation of ecological problems. *The American Naturalist* 152: 151–159.
- Grime, J.P. 1998. Benefits of plant diversity to ecosystems: immediate, filter and founder effects. *Journal of Ecology* 86: 902–910.
- Groenendijk, A.M. 1985. Ecological consequences of tidal management for the salt-marsh vegetation. *Vegetatio* 62: 415–424.
- Hammer, Ø., Harper, D.A.T. & Ryan, P.D. 2001. PAST: Paleontological Statistics Software Package for Education and Data Analysis. *Palaeontologia Electronica* 4(1): 9. Available at [http://palaeo-electronica.org/2001\\_1/past/issue1\\_01.htm](http://palaeo-electronica.org/2001_1/past/issue1_01.htm).
- Harrison, S.P., Prentice, I.C., Barboni, D., Kohfeld, K.E., Ni, J. & Sutra, J.P. 2010. Ecophysiological and bioclimatic foundations for a global plant functional classification. *Journal of Vegetation Science* 21: 300–317.
- Hooper, D.U., Chapin, F.S., Ewel, J.J., Hector, A., Inchausti, P., Lavorel, S., Lawton, J.H., Lodge, D.M., Loreau, M., Naeem, S., Schmid, B., Setälä, H., Symstad, A.J., Vandermeer, J. & Ward, D.A. 2005. Effects of biodiversity on ecosystem functioning: a consensus of current knowledge and needs for further research. *Ecological Monographs* 75: 3–35.
- Janiesch, P. 1991. Oberirdische Biomasseproduktion und Mineralstoffhaushalt von Salzwiesen der niedersächsischen Küste. *Drosera* 1/2: 127–138.
- Kazakou, E., Vile, D., Shipley, B., Gallet, C. & Garnier, E. 2006. Co-variations in litter decomposition, leaf traits and plant growth in species from a Mediterranean old-field succession. *Functional Ecology* 20: 21–30.
- Kiehl, K., Esselink, P. & Bakker, J.P. 1997. Nutrient limitation and plant species composition in temperate salt marshes. *Oecologia* 111: 325–330.
- Kingsolver, J.G. & Schemske, D.W. 1991. Path analyses of selection. *Trends in Ecology & Evolution* 6: 276–280.
- Kinzel, H. 1982. *Pflanzenökologie und Mineralstoffwechsel*. Ulmer Verlag, Stuttgart, DE.
- Knevel, I.C., Bekker, R.M., Kunzmann, D., Stadler, M. & Thompson, K. 2005. *The LEDA Traitbase – Collecting and measuring standards of life-history traits of the northwest European flora*. University of Groningen, Community and Conservation Ecology group, Groningen, NL.
- Kuijper, D.P.J., Dubbeld, J. & Bakker, J.P. 2005. Competition between two grass species with and without grazing over a productivity gradient. *Plant Ecology* 179: 237–246.
- Lavorel, S. & Garnier, E. 2002. Predicting changes in community composition and ecosystem functioning from plant traits: revisiting the Holy Grail. *Functional Ecology* 16: 545–556.
- Lavorel, S., Díaz, S., Cornelissen, J.H.C., Garnier, E., Harrison, S.P., McIntyre, S., Pausas, J.G., Pérez-Harguindeguy, N., Roumet, C. & Urcelay, C. 2007. *Plant functional types: Are we getting any closer to the holy grail? Series: global change – the IGBP series*. Springer Verlag, Berlin, DE.
- Leeuw, J.D., Olff, H. & Bakker, J.P. 1990. Year-to-year variation in peak above-ground biomass of six salt-marsh angiosperm communities as related to rainfall deficit and inundation frequency. *Aquatic Botany* 36: 139–151.
- Lenßen, G.M., Lamers, J., Stroetenga, M. & Rozema, J. 1993. Interactive effects of atmospheric CO<sub>2</sub> enrichment, salinity and flooding on growth of C<sub>3</sub> (*Elymus athericus*) and C<sub>4</sub> (*Spartina anglica*) salt marsh species. *Vegetatio* 104/105: 379–388.
- Lenßen, G.M., van Duin, W.E., Jak, P. & Rozema, J. 1995. The response of *Aster tripolium* and *Puccinellia maritima* to atmospheric carbon dioxide enrichment and their interactions with flooding and salinity. *Aquatic Botany* 50: 181–192.
- Mardia, K.V. 1970. Measures of multivariate skewness and kurtosis with applications. *Biometrika* 57: 519–530.
- Mardia, K.V. 1974. Applications of some measures of multivariate skewness and kurtosis in testing normality

- and robustness studies. *Sankhyā - The Indian Journal of Statistics Series B* 36: 115–128.
- McCarthy, M.C. & Enquist, B.J. 2007. Consistency between an allometric approach and optimal partitioning theory in global patterns of plant biomass allocation. *Functional Ecology* 21: 713–720.
- McCune, B. & Grace, J.B. 2002. Structural equation modeling. In: McCune, B. & Grace, J.B. (eds.) *Analysis of ecological communities*. pp. 233–256. MjM Software Design, Glenden Beach, OR, US.
- Mitchell, R.J. 1992. Testing evolutionary and ecological hypotheses using path analysis and structural equation modeling. *Functional Ecology* 6: 123–129.
- Moles, A.T. & Westoby, M. 2006. Seed size and plant strategy across the whole life cycle. *Oikos* 133: 91–105.
- Müller, I., Schmid, B. & Weiner, J. 2000. The effect of nutrient availability on biomass allocation patterns in 27 species of herbaceous plants. *Perspectives in Plant Ecology, Evolution and Systematics* 3/2: 115–127.
- Murphy, J. & Riley, J.P. 1962. A modified single solution method for the determination of phosphate in natural waters. *Analytica Chimica Acta* 27: 31–36.
- Naem, S. & Wright, E.J. 2003. Disentangling biodiversity effects on ecosystem functioning: deriving solutions to a seemingly insurmountable problem. *Ecology Letters* 6: 567–579.
- Niklas, K.J. 1994. *Plant allometry. The scaling of form and process*. The University of Chicago Press, Chicago, IL, US.
- Niklas, K.J. 2006. Plant allometry, leaf nitrogen and phosphorus stoichiometry, and interspecific trends in annual growth rates. *Annals of Botany* 97: 155–163.
- Niklas, K.J. & Enquist, B.J. 2003. An allometric model for seed plant reproduction. *Evolutionary Ecology Research* 5: 79–88.
- Obeso, J.R. 2002. The costs of reproduction in plants. *New Phytologist* 155: 321–348.
- Olf, H., de Leeuw, J., Bakker, J.P., Platerink, R.J., van Wijnen, H.J. & de Munck, W. 1997. Vegetation succession and herbivory in a salt marsh: changes induced by sea level rise and silt deposition along an elevation gradient. *Journal of Ecology* 85: 799–814.
- Pontes, L.D.S., Soussana, J.F., Louault, F., Andueza, D. & Carrere, P. 2007. Leaf traits affect the above-ground productivity and quality of pasture grasses. *Functional Ecology* 21: 844–853.
- Poorter, H. & de Jong, R. 1999. A comparison of specific leaf area, chemical composition and leaf construction costs of field plants from 15 habitats differing in productivity. *New Phytologist* 143: 163–176.
- Pott, R. 1995. *Farbatlas Nordseeküste und Nordseeinseln*. Ulmer Verlag, Stuttgart, DE.
- Rozema, J., Bijwaard, P., Prast, G. & Broekman, R. 1985. Ecophysiological adaptations of coastal halophytes from foredunes and salt marshes. *Vegetatio* 62: 499–521.
- Schirmer, U. & Breckle, S.W. 1982. The role of bladders for salt removal in some Chenopodiaceae (mainly *Atriplex* species). In: Sen, D.N. & Rajpurohit, K.S. (eds.) *Contributions to the ecology of halophytes*. pp. 215–231. Dr Junk, The Hague, NL.
- Schlichting, E., Blume, H.P. & Stahr, K. 1995. *Bodenkundliches praktikum*. Blackwell, Berlin, DE.
- Shipley, B. 2004. Analysing the allometry of multiple interacting traits. *Perspectives in Plant Ecology, Evolution and Systematics* 6: 235–241.
- Steward, G.R., Larher, F., Ahmand, I. & Lee, J.A. 1979. Nitrogen metabolism and salt tolerance in higher plants – halophytes. In: Jefferies, R.L. & Davy, A.J. (eds.) *Ecological processes in coastal environments*. pp. 229–241. Blackwell, Oxford, UK.
- Suding, K.N., Lavorel, S., Chapin III, F.S., Cornelissen, J.H.C., Díaz, S., Garnier, E., Goldberg, D.E., Hooper, D.U., Jackson, S.T. & Navas, M.L. 2008. Scaling environmental change through the community-level: a trait-based response-and-effect framework for plants. *Global Change Biology* 14: 1125–1140.
- SynBioSys Species Checklist 2010. Available at <http://www.synbiosys.alterra.nl/synbiosyseu/speciesviewframe.htm> (accessed: 7 June 2010)
- Tarczynski, M.C., Jensen, R.G. & Bohnert, H.J. 1993. Stress protection of transgenic tobacco by production of the osmolyte mannitol. *Science* 259: 508–510.
- Thompson, B.K., Weiner, J. & Warwick, S.I. 1991. Size-dependent reproductive output in agricultural weeds. *Canadian Journal of Botany-Revue Canadienne De Botanique* 69: 442–446.
- Tilman, D. 1988. *Plant strategies and the dynamics and structure of plant communities*. Princeton University Press, Princeton, NJ, US.
- Van Diggelen, J., Rozema, J., Dickson, D.M. & Broekman, R. 1986.  $\beta$ -3-Dimethylsulphoniopropionate, proline and quaternary ammonium compounds in *Spartina anglica* in relation to sodium chloride, nitrogen and sulphur. *New Phytologist* 103: 573–586.
- Walker, B., Kinzig, A. & Langridge, J. 1999. Plant attribute diversity, resilience, and ecosystem function: the nature and significance of dominant and minor species. *Ecosystems* 2: 95–113.
- Warton, D.I., Wright, I.J., Falster, D.S. & Westoby, M. 2006. Bivariate line-fitting methods for allometry. *Biological Reviews* 81: 259–291.
- Weiner, J., Campbell, L.G., Pino, J. & Echarte, L. 2009. The allometry of reproduction within plant populations. *Journal of Ecology* 97: 1220–1233.
- Westoby, M., Falster, D.S., Moles, A.T., Vesk, P.A. & Wright, I.J. 2002. Plant ecological strategies: some leading dimensions of variation between species. *Annual Review of Ecology and Systematics* 33: 125–159.
- Wright, I.J., Reich, P.B., Westoby, M., Ackerly, D.D., Baruch, Z., Bongers, F., Cavenders-Bares, J., Chapin, T., Cornelissen, J.H.C., Diemer, M., Flexas, J., Garnier, E., Groom, P.K., Gulias, J., Hikosaka, K., Lamont, B.B., Lee, T., Lee, W., Lusk, C., Midgley, J.J., Navas, M.-L., Niinemets, Ü., Oleksyn, J., Osada, N., Poorter, H., Poot, P., Prior, L., Pyankow, V.I., Roumet, C., Thomas, S.C., Tjoelker, M.G., Veneklaas, E.J. & Villar, R. 2004. The worldwide leaf economics spectrum. *Nature* 428: 821–827.