

relatively few taxa and/or focused on closely related species (Rothstein 1973, Sultan et al. 1998, 2001, but see Van Buskirk 2002).

Here, we investigate the relationship between environmental and phenotypic variation in coastal salt marsh plants. Coastal salt marshes are ideal systems for examining this relationship because they contain severe environmental gradients that have been correlated to variation in plant phenotype (Valiela et al. 1978, Seliskar 1985a, 1985b, 1987, Bertness and Ellison 1987). While some salt marsh plant species occur across a broad range of environmental variables, others are more restricted in distribution (Bertness et al. 1992, Gough and Grace 1998, Sanchez et al. 1998, Pan et al. 1998, Rand 2000). Previous studies of intraspecific variation in salt marsh plant phenotypes have primarily focused on differences between extreme height forms, especially for the grass *Spartina alterniflora* (Anderson and Treshow 1980, Gallagher et al. 1988, Trnka and Zedler 2000), or differences between isolated populations (Silander 1979, 1984, 1985, Silander and Antonovics 1979, Hester et al. 1996, Hamilton 1997). However, none of these studies investigated whether habitat breadth corresponded to phenotypic variability across a larger pool of species. Although this idea could be tested on many spatial scales ranging from the local to the geographic, we address it at the local scale of cross-marsh gradients, because local environmental gradients in salt marshes are so strong.

This study documents the extent of aboveground phenotypic variation for the twelve most common plant species that occur in Georgia salt marshes, and correlates the observed phenotypic variation with several environmental variables. We test the hypotheses that 1) within species, plant traits correlate with environmental variables, and 2) species with wider environmental ranges have more variable phenotypes.

Methods

Study sites and species

We studied twelve plant species that are common in southeastern USA salt marshes and represent six families (Asteraceae: *Aster tenuifolius* L., *Borrchia frutescens* L., *Iva frutescens* L.; Bataceae: *Batis maritima* L.; Chenopodiaceae: *Salicornia bigelovii* Torrey, *Salicornia virginica* L.; Juncaceae: *Juncus roemerianus* Scheele; Plumbaginaceae: *Limonium*

carolinianum (Walt.) Britt.; Poaceae: *Distichlis spicata* (L.) Greene, *Spartina alterniflora* Loisel., *Spartina patens* (Aiton) Muhl., *Sporobolus virginicus* (L.) Kunth; all nomenclature follows Radford et al. 1968). We worked at seventeen sites on Sapelo Island, Georgia, USA (31° 28'N, 81° 14'W). The vegetation patterns in Sapelo Island marshes are typical of southeastern marshes in the United States (Pomeroy and Wiegert 1981). Lower elevations of the marsh are subject to daily tidal submergence and are dominated by *Spartina alterniflora*. The higher elevations of the marsh are flooded irregularly and are often characterized by highly saline salt pans and associated salt-tolerant species such as *Salicornia virginica*, *Salicornia bigelovii*, *Batis maritima*, *Borrchia frutescens*, *Distichlis spicata* and *Sporobolus virginicus* (Antlfinger 1981). The terrestrial border of the marsh is typically dominated by *Juncus roemerianus*, *Spartina patens* or *Iva frutescens*. *Aster tenuifolius* and *Limonium carolinianum* occur at higher elevations mixed in with the zonal dominants. The details of the plant zonation patterns vary from site to site, and not every species occurs at every marsh site. The twelve species that we studied represent the vast majority of the species and the plant biomass present at all of our sites.

Phenotypic and environmental sampling

We sampled plant traits between 1 July and 16 August, 1999 ($N = 1057$ plants). Within this time frame, each species was sampled after it had flowered and completed the majority of its vegetative growth across the marsh environmental gradients. Each species was sampled along one transect at each of eight or nine sites. Because the species composition of each site varied, the number of species sampled per site ranged from one to twelve. Individual transects ran from the upper to the lower elevational range of the target species at each site. Ten plants were selected along each transect using a stratified-random sampling scheme to ensure representation of the full extent of environmental breadth. Due to the broad horizontal and elevation range of *Spartina alterniflora*, we collected data on twenty individuals at each of the eight sites for this species.

Traits measured for each species included plant height, number of leaves, length, width and thickness of three fully emerged leaves, length of the third internode, and other traits as appropriate for the growth form of each plant species (Table 1). Height in the four grasses and most of the forbs and small

Table 1. Traits measured for each plant species. For each species, all traits indicated were used in a Principal Components Analysis (PCA). PCA1 indicates the amount of variation in all traits explained by the first principal component axis. Ht = height, LV = leaf volume, PD = plant depth, PW = plant width, LS = leaf serration, BD = base diameter, BP = number of primary branches, BS = number of secondary branches.

	Traits measured											PCA1
	Ht	Leaf number	Leaf size	Leaf length	Leaf width	Leaf thickness	Stem diameter	Internode length	Other			
<i>Aster tenuifolius</i>	X	X	X	X	X		X	X				0.54
<i>Batis maritima</i>	X	X	X	X	X	X			LV			0.52
<i>Borrchia frutescens</i>	X	X	X	X	X	X						0.40
<i>Distichlis spicata</i>	X	X	X	X	X		X	X				0.47
<i>Iva frutescens</i>	X	X	X	X	X	X			PD, PW, LS (1-4)			0.34
<i>Juncus roemerianus</i>	X	X	X	X	X							0.63
<i>Limonium carolinianum</i>	X	X	X	X	X		X		BD			0.68
<i>Salicornia bigelovii</i>	X	X	X	X	X		X		PW, BP, BS			0.49
<i>Salicornia virginica</i>	X	X	X	X	X		X		BP, BS			0.64
<i>Spartina alterniflora</i>	X	X	X	X	X			X				0.62
<i>Spartina patens</i>	X	X	X	X	X		X	X				0.58
<i>Sporobolus virginicus</i>	X	X	X	X	X		X	X				0.45

shrubs was measured from the ground to the uppermost leaf node on the stem. The exceptions include *Iva* for which height was measured to the top of the canopy, the *Salicornia* species for which height was measured to the top of the uppermost appressed leaf, and *Juncus* for which height was measured to the end of each needle-like leaf. For flowering individuals of *Borrchia* and *Aster*, height was measured to the uppermost flower head. Leaf size was estimated as maximum length \times maximum width in all plants with the exception of the succulent *Batis* for which leaf volume was estimated by leaf size \times maximum thickness. Stem diameter was measured at the base of the stem with calipers. Base width was measured below the basal rosette (*Limonium*). Leaf serration was scored (*Iva*) on a scale of one to four with one indicating smooth leaves and four indicating highly serrated leaves.

After all plants were tagged and traits measured, we collected a soil sample adjacent to each plant within a three day period (21–23 August 1999). Soil was dried (60 °C to constant weight) to determine relative water content, rehydrated in a known volume of distilled water to determine original pore water salinity (Pennings and Richards 1998), and ashed (550 °C for 12 hours) to determine organic content. We surveyed the soil elevation at the base of each tagged plant to the nearest millimeter. Elevation was converted to a relative index for each species at each site by setting the lowest value for the species to 0 and the highest to 1. We chose these four environmental variables because 1) salinity is very important, 2) water content of soil, given the broad range of values obtained here, is correlated with redox and oxygen availability, which are much harder to measure, and 3) organic content should correlate with nutrient availability and 4) elevation represents a combination of environmental factors. In addition, it was logistically feasible to sample these four environmental variables for large number of plants in a short time period that minimized temporal variation.

Statistical analysis

Intraspecific relationships. Plant traits were regressed against environmental variables using simple linear and multiple regressions in SAS (SAS 2000). To simplify cross species comparisons, we report here the results for the three most-commonly measured traits (plant height, leaf size and number of leaves) and a composite variable (the first principal component axis

obtained from a PC ordination of all of the traits measured for each species). Height and leaf number for all species were natural log transformed and leaf size was untransformed for most species with the exception of *Spartina alterniflora* and *Limonium* for which leaf size was Box Cox transformed (JMP 1999) to meet the assumptions of normality and homoscedasticity. Soil proportion water content and ash content were arcsine-square root transformed and relative elevation was Box Cox transformed (JMP 1999) to meet regression assumptions. This approach uses some of the data in more than one statistical test for each species. Given that the number of multiple tests is small, we did not adjust P-values.

We ran full multiple regression models with the four environmental variables for each dependent variable. After removing environmental variables that were not at least marginally significant ($\alpha > 0.10$), we ran the models again to determine if marginally significant variables became significant in reduced models. Final reduced models include only those variables significant at the $\alpha < 0.05$ level. For each species, we ran a separate principal components analysis (PCA) which included all of the traits measured for that species (Table 1) using proc princomp in SAS (SAS 2000). All traits entered into the analysis were left unstandardized. The PCA1 accounted for between 34–68% of the variation in each of the species (Table 1). PCA1 was consistently evenly loaded across three to five plant traits and no species showed a strong correlation between PCA1 and any one trait.

Interspecific relationships. To quantify the extent of phenotypic variation, we calculated the coefficient of variation, ($CV = \text{standard deviation} / \text{mean}$). We used the CV of plant traits to account for the fact that, as is typical of many ecological data sets, the variance of traits rose with their mean. Thus, the variance in most traits of the larger plant species was greater than the variance of the smaller species. In this case, calculating the CV provides a measure of variation that is less affected by the mean. To quantify the range of each environmental variable, we calculated $2 \times$ the standard deviation of that variable (using the CV of environmental variables in analyses did not alter our conclusions). To examine the relationship between phenotypic variation and habitat breadth among species ($N = 12$), we regressed the CVs of height and leaf size on the range of each environmental variable (elevation was not used in these analyses because it was a relative index standardized to one). We also examined the relationship between a

composite phenotypic variation variable (PCA1 of the three phenotypic CV values in Table 2) and a composite environmental variation variable (PCA1 of the three environmental ranges in Table 2) for the 10 species for which all of these variables were measured.

Results

Extent of phenotypic and environmental variation

Plant species differed considerably in phenotypic variability (Table 2). The coefficient of variation of plant height ranged from 0.24 (*Juncus*) to 0.64 (*Salicornia virginica*). The CV of leaf size ranged from 0.22 (*Salicornia bigelovii*) to 0.82 (*Limonium*). There was a much broader range in the CV of the number of leaves, which ranged from 0.28 (*Spartina patens*) to 1.92 (*Salicornia bigelovii*).

Similarly, the range of environmental variables differed considerably among plant species (Table 2). The range ($2 \times SD$) of salinity varied 4-fold among species, from 27.7 (*Iva*) to 120.8 (*Salicornia bigelovii*). The range of water content varied 3-fold, from 0.10 (*Salicornia bigelovii*) to 0.32 (*Juncus*). The range of organic content varied 3-fold, from 0.05 (*Limonium*) to 0.15 (*Iva*).

Intraspecific relationships: environmental variation and plant phenotype

For each plant species, variation in plant phenotype was correlated with variation in the environment, and over $\frac{3}{4}$ of the relationships between height, leaf size or leaf number and environmental traits were significant (Table 3). There was variation among traits and species in the combination of environmental variables that predicted plant traits. However, salinity was the most common predictor variable, followed by organic content, water content and elevation. When multiple environmental variables were significant predictors of plant traits, salinity usually entered first into the stepwise regression model. As expected, the relationship between plant traits and 1) salinity was almost always (28/30) negative, 2) organic content was almost always (20/22) positive, 3) water content was usually (10/14) negative, and 4) elevation was usually (7/10) positive.

In all but three species, height correlated with environmental variables more strongly than did any

Table 2. Variation in three plant traits (CV) and mean ($2 \times$ SD) of three environmental variables for the twelve plant species. NM = this variable was not measured for this species.

Species	Height (CV)	Average leaf size (CV)	Number of leaves (CV)	Phenotypic PCA1	Salinity mean ($2 \times$ SD) ppt	Proportion soil water content mean ($2 \times$ SD) g/g	Proportion soil organic content mean ($2 \times$ SD) g/g	Environmental PCA1
<i>Aster tenuifolius</i>	0.53	0.80	0.52	-1.15111	43.8 (44.9)	0.26 (0.22)	0.04 (0.06)	-0.2061
<i>Batis maritima</i>	0.53	0.23	1.29	1.638959	67.1 (68.2)	0.27 (0.18)	0.05 (0.08)	-0.67694
<i>Borrichia frutescens</i>	0.46	0.50	0.86	0.029147	54.6 (54.3)	0.24 (0.17)	0.05 (0.08)	-0.49835
<i>Distichlis spicata</i>	0.35	0.40	0.39	-0.50632	64.6 (68.4)	0.32 (0.26)	0.08 (0.13)	1.15901
<i>Iva frutescens</i>	0.29	0.36	NM		39.4 (27.7)	0.24 (0.22)	0.08 (0.15)	
<i>Juncus roemerianus</i>	0.24	0.46	0.40	-0.97409	43.8 (28.7)	0.36 (0.32)	0.08 (0.13)	2.574508
<i>Limonium carolinianum</i>	NM	0.82	0.33		44.9 (51.0)	0.23 (0.19)	0.03 (0.05)	
<i>Salicornia bigelovii</i>	0.33	0.22	1.92	1.854019	106.8 (120.8)	0.20 (0.10)	0.03 (0.07)	-2.76196
<i>Salicornia virginica</i>	0.64	0.52	1.78	1.555112	65.2 (72.8)	0.30 (0.23)	0.07 (0.11)	0.354563
<i>Spartina alterniflora</i>	0.47	0.56	0.32	-0.77625	51.7 (48.3)	0.59 (0.24)	0.15 (0.09)	0.542943
<i>Spartina patens</i>	0.29	0.65	0.28	-1.61312	32.8 (39.7)	0.28 (0.23)	0.08 (0.12)	1.227726
<i>Sporobolus virginicus</i>	0.48	0.40	0.47	-0.05635	58.3 (94.3)	0.23 (0.15)	0.04 (0.07)	-1.71539

Table 3. Multiple regression models for 3 phenotypic traits and PCA1 for the 12 plant species. Environmental variables are listed in the order in which they loaded into a stepwise regression. The sign of the coefficient for each environmental variable is indicated. The plant trait with the best fit for each species is indicated in bold. * = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$. S = soil salinity, H = proportion soil water content, O = proportion soil organic content, E = elevation, NS = not significant, NM = this variable not measured for this species.

Species	Ln height		Leaf size		Ln leaf number		PCA 1	
	R ²	predictors	R ²	predictors	R ²	predictors	R ²	predictors
<i>Aster tenuifolius</i>	0.49 ***	+O,-H,-S,-E	NS		0.16 ***	+O	0.22 **	-S,-H,+O
<i>Batis maritima</i>	0.26 ***	-S	NS		0.17 **	-S	NS	
<i>Borrichia frutescens</i>	0.20 ***	-S,+O	0.22 ***	-S,+E	0.19 ***	-S,+O,-H	0.24 ***	-S,+E
<i>Distichlis spicata</i>	0.65 ***	+H,-S	0.32 ***	+O,-H,-S	0.18 ***	+O	0.47 ***	+O
<i>Iva frutescens</i>	0.17 **	+E	NS		NM		NS	
<i>Juncus roemerianus</i>	0.50 ***	-S	0.45 ***	+H,-S,+E,-O	NS		0.48 ***	-S,+H,+E,-O
<i>Limonium carolinianum</i>	NM		0.25 ***	-S,+O,-H	0.30 ***	-S,+O	0.23 ***	-S,+O
<i>Salicornia bigelovii</i>	0.10 **	+O	0.33 ***	+O	0.17 **	+O,+S	0.30 ***	+O
<i>Salicornia virginica</i>	0.71 ***	-S,+O,-H	0.32 ***	-S,-H	0.41 ***	-S,+O,-H	0.57 ***	-S,+O,-H
<i>Spartina alterniflora</i>	0.53 ***	-S,-E	0.31 ***	-S,+E	0.17 ***	-S,-E	0.34 ***	-S
<i>Spartina patens</i>	0.35 ***	-S,+H	0.31 ***	-S,+E	0.11 **	+S	0.26 ***	-S
<i>Sporobolus virginicus</i>	0.33 ***	-S,+O	NS		0.13 *	+O,-H	0.06 *	

other trait or PCA1. The model for leaf size showed the best fit for *Salicornia bigelovii* and the model for leaf number showed the best fit for *Limonium*. Surprisingly, the model for the composite variable PCA1 showed the best fit in only one case (*Borrchia*), and in this case the R^2 for PCA1 (0.24) was only slightly greater than the R^2 for leaf size (0.22).

Interspecific relationships: phenotypic variation and habitat breadth

Linear regressions of plant trait CVs on range ($2 \times$ SD) of environmental variables were not significant (Figure 1) and did not support the hypothesis that species with wider environmental ranges would have more variable phenotypes. However, quadratic regressions revealed that the relationship between CV height and range of salinity was significant ($P = 0.006$, $R^2 = 0.65$). This relationship indicated that species inhabiting intermediate ranges of salinity exhibited the most variation in height whereas species inhabiting extremely small or extremely large ranges in salinity had less variation in height. Other quadratic regressions of plant trait CVs on range of environmental variables were not significant.

The PCA1 of the principal component analysis on the three phenotypic CVs accounted for 52.9% of the variation in the three traits with high loadings on leaf size (+) and leaf number (-). The PCA1 of the principal component analysis on the three environmental ranges accounted for 78.6% of the variation in the three variables with high loadings on all three variables: salinity (-), water content (+) and organic content (+). Regression of the composite phenotypic PCA1 on the composite environmental PCA1 yielded a marginally significant ($P = 0.054$) negative relationship (Figure 2).

Discussion

Salt marshes contain steep environmental gradients: conditions are fairly mild near the terrestrial border of the marsh but become so severe in salt pans and extremely waterlogged areas that even the most highly-adapted salt marsh plants cannot survive (Pennings and Bertness 2001). Across these strong environmental gradients, phenotypic variation of plants was correlated with environmental variables, as predicted by our first hypothesis. In contrast, our second hypothesis was not supported by linear comparisons

of single plant traits with single environmental variables. We found instead that variation in height was maximized in species with intermediate ranges of salinity. In addition, a composite, complex phenotypic response (phenotypic PCA1) appeared to be related to a composite, complex environmental variable (environmental PCA1). This relationship suggests that species increase variation in the number of leaves, but decrease variation in leaf size in response to an increase in range of salinity and a decrease in range of water and organic content. Thus, there is a relationship between environmental and phenotypic variation, as we hypothesized, but the nature of this relationship is quite complex.

Intraspecific relationships: environmental variation and plant phenotype

All twelve of the salt marsh plant species that we studied displayed substantial variation in phenotype. Most of the previous attention paid to phenotypic variation in salt marsh plants has focused on intraspecific variation in *Spartina alterniflora*, the most abundant and widespread salt marsh plant on the Atlantic Coast of the United States (Valiela et al. 1978, Pomeroy and Wiegert 1981), although intraspecific variation has also been documented in some other salt marsh plant species (Antlfinger 1981, Seliskar 1985a, 1985b, 1987). Here, we show that intraspecific phenotypic variation is a general phenomenon of 12 common southeastern USA salt marsh plants. Given the strong environmental gradients present in salt marsh habitats, marked intraspecific variation in height and other phenotypic traits is probably the rule for all species of salt marsh plants. For the few salt marsh plant species that have been studied, phenotypic variation is due largely to phenotypic plasticity (Valiela et al. 1978, Anderson and Treshow 1980, Antlfinger 1981, Seliskar 1985b, Richards et al. in prep.), although genetic differentiation can also play an important role (Antlfinger 1981, Silander 1985, Gallagher et al. 1988, Proffitt et al. 2003). Further studies are required to determine the relative contributions of plasticity and genetic differentiation to determining phenotypic variation in the particular species that we studied.

The majority of the relationships between plant traits and environmental variables were significant, indicating that variation in plant phenotype is predictable and correlated with environmental variation. For several plant species, some combination of

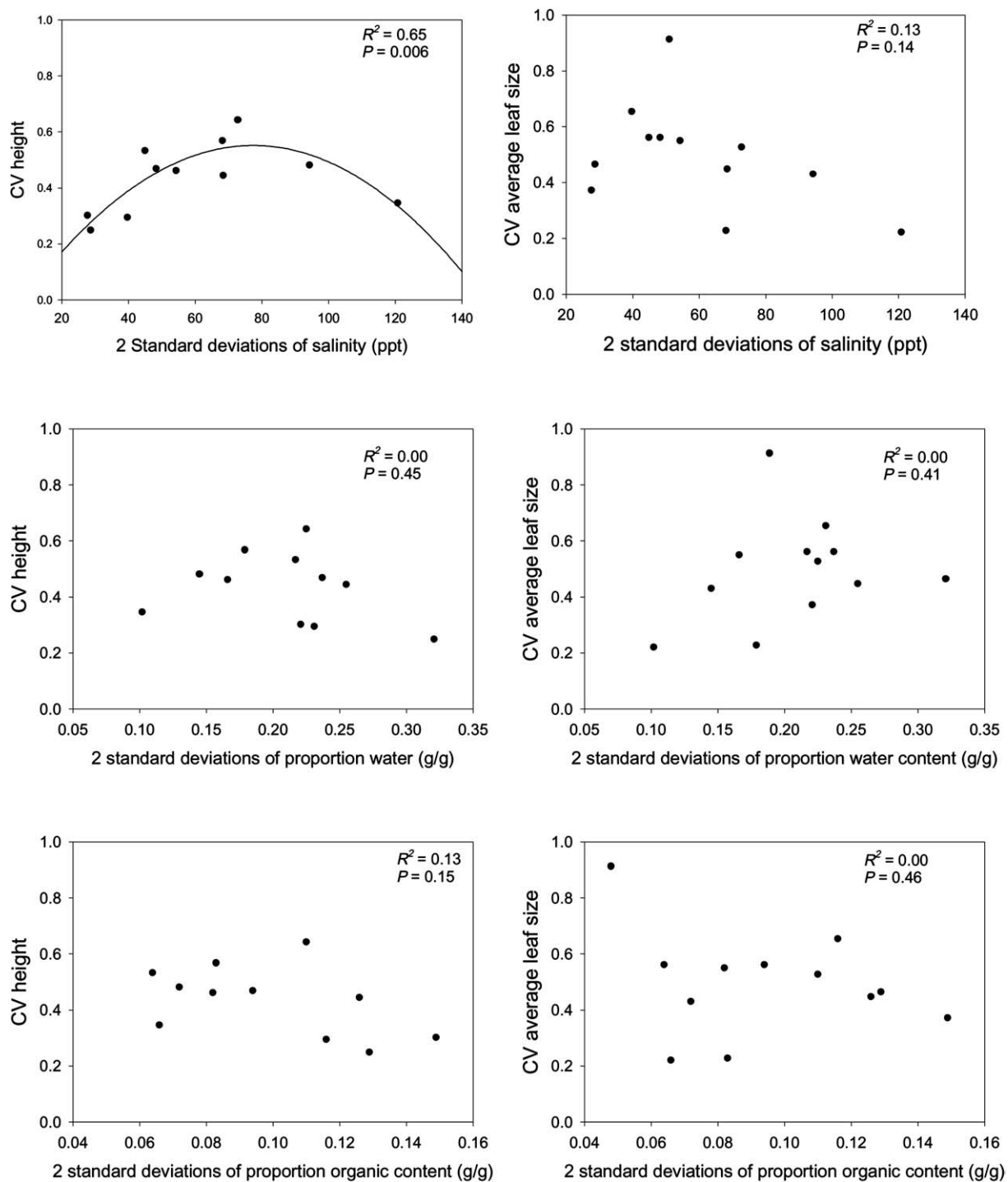


Figure 1. Relationship between CV of height or leaf size and range ($2 \times \text{SD}$) of soil variables. Each point represents a single plant species. Adjusted R^2 and P - values are shown.

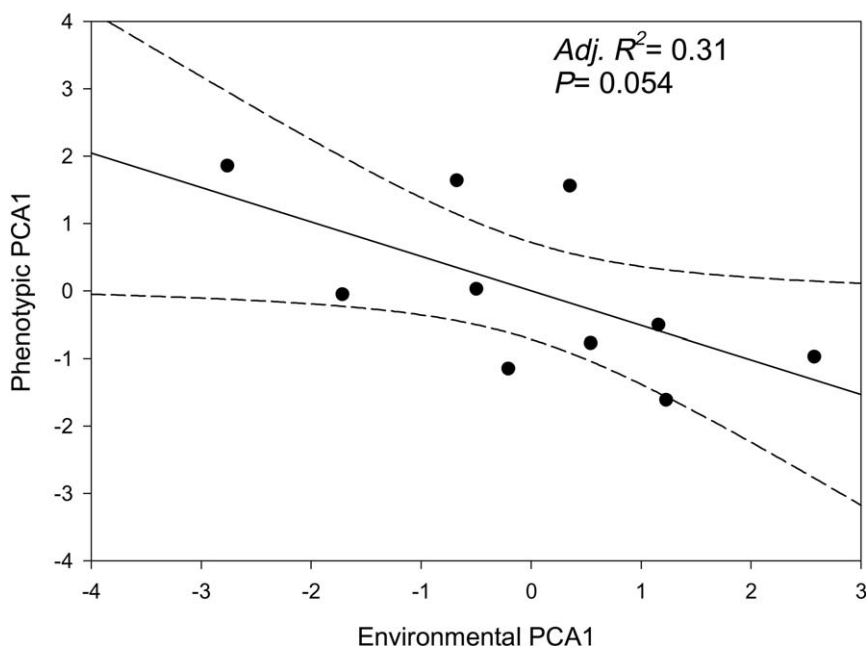


Figure 2. Relationship between phenotypic PCA1 (combining CV's for height, leaf size and number of leaves) and environmental PCA1 (combining $2 \times$ standard deviation of soil salinity, proportion soil water content and proportion soil organic content). Data includes only those 10 species for which all three phenotypic traits were measured (excluding *Iva* and *Limonium*).

the four environmental variables explained $> 45\%$ of the variation in one or more of the traits (Table 3: *Aster*, *Distichlis*, *Juncus*, *Salicornia virginica*, *Spartina alterniflora*). In the worst case, environmental variables explained only 16% of the variation in height of *Iva*, and did not predict *Iva* leaf size. However, we sampled environmental traits on only one date, and it is likely that an average of environment across seasons and tidal conditions would have explained more of the phenotypic variation in these species. Alternatively, phenotypic variation in these species may be better explained by other environmental variables that we did not measure.

The negative relationships that we observed between plant traits and salinity and waterlogging are consistent with the known physiological costs imposed on plants by these variables (Ponnamperuma 1972, Flowers 1977, Mendelssohn and Morris 2000). The high frequency with which organic content entered into the regressions suggests that, despite strong stress gradients in salt marshes, plant productivity may also be mediated by soil quality. Soils in these marshes had low organic content (averaging $< 8\%$ for the species with positive relationships with organic content), and under these conditions organic content may reflect the availability of a wide variety

of nutrients (Lindau and Hossner 1981, Craft et al. 1991, Padgett and Brown 1999). Although salinity, waterlogging and organic content all vary across elevation, elevation *per se* was rarely a significant predictor of plant traits, likely because the stresses imposed on plants by these variables may interact in complex ways across the elevation gradient (Mendelssohn and Morris 2000, Proffitt et al. 2003).

Because many studies have argued that fitness is not related to any one single trait, but rather to a suite of traits and their interactions (Clausen et al. 1948, Lechowicz 1984, Chapin et al. 1993), we expected that PCA1 would offer a composite 'plant phenotype' that might give the best overall indication of plant response to environmental variables. However, we found that PCA1 correlated with environmental variables better than single plant traits for only one species (*Borrchia*). In most cases, plant height correlated with environmental variables better than any other plant trait or PCA1. Because height correlates with biomass for plants in general, and many of these species in particular (Bertness and Ellison 1987, Pennings and Callaway 2000), these results suggest plant fitness may also vary across salt marsh environmental gradients. Because most of these species are clonal

perennials, however, documenting this variation in fitness will be a challenging task.

Interspecific relationships: phenotypic variation and habitat breadth

Ecologists have long speculated that species with wider environmental ranges would have broader ranges in phenotype (Van Valen 1965, Baker 1974, Sultan 2001). Most of the tests of this hypothesis, however, have involved small numbers of species and/or closely related taxa (Rothstein 1973, Sultan et al. 1998, Sultan 2001). In one study that did examine variation across taxa, Van Buskirk (2002) found that frog species with the widest habitat ranges showed the largest morphological responses to predator variation. In contrast, we found that for plants from 6 families, linear regressions of plant trait CVs on ranges in soil variables ($2 \times$ standard deviations) were not significant in any case ($P > 0.05$, Figure 1). Thus our data do not support the hypothesis that species occupying broad environmental ranges will have a linear response in breadth of phenotypes. We did, however, find a significant quadratic relationship between height CV and range of salinity suggesting that species that inhabit areas with intermediate ranges of salinity exhibit the most variation in height whereas species inhabiting extremely small or extremely large ranges of salinity have less variation in height. This trend is consistent with environmental canalization (Debat and David 2001, Wagner et al. 1997) for an optimum height in species that inhabit the most extreme range in salinities which is relaxed in species that inhabit intermediate ranges of salinities. However, the quadratic nature of this relationship is mostly influenced by the data for *Salicornia bigelovii* which inhabits the broadest range of salinity (2 SD = 120.8 ppt) and has very little phenotypic response to the salinity gradient (Table 3).

The lack of a linear relationship between phenotypic and environmental range in our study is likely due to distantly-related taxa responding to environmental challenges in different ways. For example, there are several different physiological and morphological solutions to the problem of coping with high salt environments (Hasegawa et al. 2000, Flowers et al. 1977). In contrast, closely-related taxa are likely to use the same mechanisms to respond to similar challenges. Thus, studies of closely-related taxa are more likely to observe a positive correlation between phenotypic and environmental range (Rothstein 1973,

Sultan 1998, 2001, but see Van Buskirk 2002). Alternatively, it may be that, environmental gradients in the field, and the responses of organisms to these gradients, are inherently complex and multivariate. For this group of species, we found that a composite, complex phenotypic response (phenotypic PCA1) explained 53% of the variance in the height, number and size of leaves. This composite variable was not significantly correlated with variation in height, but rather represented an increase in variation in the number of leaves and a corresponding decrease in variation in leaf size. The composite, complex environmental variable represented an increase in range of salinity inhabited with a corresponding decrease in range of water and organic content. The regression of these two composite variables suggests that the relationship between phenotypic variation and environmental variation is not simply linear. Instead, it appears that species increase variation in the number of leaves, but decrease variation in leaf size, in response to an increase in range of salinity and decrease in range of water and organic content.

Our results come with two caveats. First, our study was observational rather than experimental. It is possible that biotic interactions or other factors could have obscured additional relationships between phenotypic and environmental variation that might have been revealed by an experimental manipulation. In particular, some of the plant species are competitively excluded from habitats that they are physiologically capable of inhabiting (Pennings and Bertness 2001), thereby limiting the range of environmental variation that we observed. Second, given the modest number of species studied (12), the results of our species-level regressions may be influenced by single points. In particular, the linear relationship between the CV of height and the range of salinity (Figure 1 top left) would be significant ($R^2 = 0.48$, $P = 0.03$) if one data point (*Salicornia bigelovii*) were removed.

In sum, we found that variability in phenotypes of all twelve salt marsh plants was correlated with variation in the physical environment; however, linear, univariate relationships between the range of environments occupied by a species and the range of variation in phenotype did not occur. Rather, the relationship between variation in phenotype and variation in the environment was non-linear and/or multivariate in nature. We conclude that linear relationships between environmental and phenotypic variation are most likely to be found when comparing closely-related taxa and simple gradients than when comparing a

broad range of taxa across complex gradients in the field. In the field, environmental gradients, and the responses of organisms to these gradients, are likely to be complex and multivariate.

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References

- Anderson C.E. and Treshow M. 1980. A review of environmental and genetic factors that effect *Spartina alterniflora* height forms. *Estuaries* 3: 168–176.
- Antlfinger A.E. 1981. The genetic basis of microdifferentiation in natural and experimental populations of *Borrichia frutescens* in relation to salinity. *Evolution* 35: 1056–1068.
- Baker H.G. 1974. The evolution of weeds. *Annual Review of Ecology and Systematics* 5: 1–24.
- Bertness M.D. and Ellison A.M. 1987. Determinants of pattern in a New England salt marsh plant community. *Ecological Monographs* 57: 129–147.
- Bertness M.D., Gough L. and Shumway S.W. 1992. Salt tolerances and the distribution of fugitive salt marsh plants. *Ecology* 73: 1842–1851.
- Briggs D. and Walters S.M. 1997. *Plant Variation and Evolution*. 3rd edn . Cambridge University Press, Cambridge, UK.
- Chapin F.S. III, Autumn K. and Pugnaire F. 1993. Evolution of suites of traits in response to environmental stress. *American Naturalist* 142: S78–S92.
- Clausen J.D., Keck D. and Heisey W.M. 1948. Experimental studies on the nature of species. III. Environmental responses of climatic races of *Achillea*. Carnegie Institution of Washington Publication 520.
- Craft C.B., Seneca E.D. and Broome S.W. 1991. Porewater chemistry of natural and created marsh soils. *Journal of Experimental Marine Biology and Ecology* 152: 187–200.
- Debat V. and David P. 2001. Mapping phenotypes: canalization, plasticity and developmental stability. *Trends in Ecology and Evolution* 16: 555–561.
- Flowers T.J., Troke P.F. and Yeo A.R. 1977. The mechanism of salt tolerance in halophytes. *Annual Review of Plant Physiology* 28: 89–121.
- Gallagher J.L., Somers G.F., Grant D.M. and Seliskar D.M. 1988. Persistent differences in two forms of *Spartina alterniflora*: a common garden experiment. *Ecology* 69: 1005–1008.
- Gough L. and Grace J.B. 1998. Effects of flooding, salinity and herbivory on coastal plant communities, Louisiana, United States. *Oecologia* 117: 527–535.
- Hamilton M.B. 1997. Genetic fingerprint-inferred population subdivision and spatial genetic tests for isolation by distance and adaptation in the coastal plant *Limonium carolinianum*. *Evolution* 51: 1457–1468.
- Hasegawa P.M., Bressan R.A., Zhu J.- K. and Bohnert H.J. 2000. Plant cellular and molecular responses to high salinity. *Annual Review of Plant Physiology* 51: 463–499.
- Hester M.W., Mendelssohn I.A. and McKee K.L. 1996. Intraspecific variation in salt tolerance and morphology in the coastal grass *Spartina patens* (Poaceae). *American Journal of Botany* 83: 1521–1527.
- Lindau C.W. and Hossner L.R. 1981. Substrate characterization of an experimental marsh and three natural marshes. *Soil Science Society of America Journal* 45: 1171–1176.
- Lechowicz M.J. 1984. The effects of individual variation in physiological and morphological traits on the reproductive capacity of the common cocklebur, *Xanthium strumarium* L. *Evolution* 38: 833–844.
- Mendelssohn I.A. and Morris J.T. 2000. Eco-physiological controls on the productivity of *Spartina alterniflora* Loisel.. In: *Concepts and Controversies in Tidal Marsh Ecology*. (eds. Weinstein M.P. and Kreeger D.A. Kluwer Academic Publishers.
- Padgett D.E. and Brown J.L. 1999. Effects of drainage and soil organic content on growth of *Spartina alterniflora* (Poaceae) in an artificial salt marsh mesocosm. *American Journal of Botany* 86: 697–702.
- Pan D., Bouchard A., Legendre P. and Domon G. 1998. Influence of edaphic factors on the spatial structure of inland halophytic communities: a case study in China. *Journal of Vegetation Science* 9: 797–804.
- Pennings S.C. and Bertness M.D. 2001. Salt marsh communities.. In: *Marine Community Ecology* (eds. Bertness M.D., Gaines S.D. and Hay M.E.). Sinauer Associates.
- Pennings S.C. and Callaway R.M. 2000. The advantages of clonal integration under different ecological conditions: a community-wide test. *Ecology* 81: 709–716.
- Pennings S.C. and Richards C.L. 1998. Effects of wrack burial in salt- stressed habitats: *Batis maritima* in a southwest Atlantic salt marsh. *Ecography* 21: 630–638.
- Pomeroy L.R. and Wiegert R.G. 1981. *The Ecology of a Salt Marsh*. Springer-Verlag, New York.
- Ponnamperuma F.N. 1972. The chemistry of submerged soils. *Advances in Agronomy* 24: 29–95.
- Proffit C.E., Travis S.E. and Edwards K.R. 2003. Genotype and elevation influence *Spartina alterniflora* colonization and growth in a created salt marsh. *Ecological Applications* 13: 180–192.
- Radford A.E., Ahles H.E. and Bell C.R. 1968. *Manual of the Vascular Flora of the Carolinas*. The University of North Carolina Press, Chapel Hill.
- Rand T. 2000. Seed supply, habitat suitability and the distribution of halophytes across a New England salt marsh. *Journal of Ecology* 88: 608–621.
- Reznick D. and Travis J. 1996. The empirical study of adaptation in natural populations.. In: *Adaptation*. (eds. Rose M.R. and Lauder G.V.), Academic Press, New York.

- Rothstein S.I. 1973. The niche-variation model- is it valid? *American Naturalist* 107(957): 598–620.
- Sanchez J.M., Otero X.L. and Izco J. 1998. Relationships between vegetation and environmental characteristics in a salt-marsh system on the coast of Northwest Spain. *Plant Ecology* 136: 1–8.
- Seliskar D.M. 1985a. Morphometric variations of five tidal marsh halophytes along environmental gradients. *American Journal of Botany* 72: 1340–1352.
- Seliskar D.M. 1985b. Effect of reciprocal transplanting between extremes of plant zones on morphometric plasticity of five plant species in an Oregon salt marsh. *Canadian Journal of Botany* 63: 2254–2262.
- Seliskar D.M. 1987. The effects of soil moisture on structural and biomass characteristics of four salt marsh plants. *Journal of Experimental Botany* 38: 1193–1202.
- Silander J.A. 1979. Microevolution and clone structure in *Spartina patens*. *Science* 203: 658–660.
- Silander J.A. 1984. The genetic basis of the ecological amplitude of *Spartina patens*. III. Allozyme variation. *Botanical Gazette* 145: 569–577.
- Silander J.A. 1985. The genetic basis of the ecological amplitude of *Spartina patens*. II. Variance and correlation analysis. *Evolution* 39: 1034–1052.
- Silander J.A. and Antonovics J. 1979. The genetic basis of the ecological amplitude of *Spartina patens*. I. Morphometric and physiological traits. *Evolution* 33: 1114–1127.
- Sultan S.E. 1995. Phenotypic plasticity and plant adaptation. *Acta Botanica Neerlandica* 44: 363–383.
- Sultan S.E., Wilczek A.M., Bell D.L. and Hand G. 1998. Contrasting ecological breadth of co-occurring annual *Polygonum* species. *Journal of Ecology* 86: 363–383.
- Sultan S.E. 2001. Phenotypic plasticity for fitness components in *Polygonum* species of contrasting ecological breadth. *Ecology* 82: 328–343.
- Trnka S. and Zedler J.B. 2000. Site conditions, not parental phenotype, determine the height of *Spartina foliosa*. *Estuaries* 23(4): 572–582.
- Valiela I., Teal J.M. and Deuser W.G. 1978. The nature of growth forms in the salt marsh grass *Spartina alterniflora*. *American Naturalist* 112: 461–470.
- Van Buskirk J. 2002. A comparative test of the adaptive plasticity hypothesis: relationships between habitat and phenotype in anuran larvae. *American Naturalist* 160: 87–102.
- Van Valen L. 1965. Morphological variation and width of ecological niche. *American Naturalist* 99: 377–390.
- Wagner G.P., Booth G. and Bagheri-Chaichian H. 1997. A population genetic theory of canalization. *Evolution* 51: 329–347.