

Changes in forest structure along an elevational gradient in the Peruvian Andes cause species-specific stress responses in tree seedlings

Abstract

4 bullet points (1) research conducted + rationale, (2) central methods, (3) key results, (4) main conclusions including key points of discussion.

Introduction

Rapid anthropogenic climate change is causing many species, across a wide range of taxa, to shift their distributions in space (???). ? estimates average latitudinal and poleward migration rates of NUM and NUM, respectively. For sessile taxa such as trees, range shifts occur as a result of differential recruitment and mortality over space, at the leading and trailing edges of their range REF. Previous studies have suggested that the ability of tree species to respond to changes in mean annual temperature and precipitation regime will be important in determining species success over the coming century (???). Species responses may occur either in the form of adaptation, *i.e.* changes in phenology, physiology and morphology, or through range shifts over space as species track environmental conditions (Bellard et al. 2012). Range shifts of plant species have been observed in many studies across the world REF, while the number of studies documenting adaptational responses are few, potentially indicating that climate change is occurring so rapidly as to prevent effective adaptational responses. Predicting range shifts and more importantly the ability for species to shift their ranges across space has become an active field of research REFS FROM BELLARD 2012. Understanding species range shifts can help conservation scientists to identify species and species assemblages at risk of extinction and can inform strategies to mitigate the effects of climate change on biodiversity.

The majority of efforts to predict future species range shifts as a conservation tool have used bioclimatic envelope models (?). Bioclimatic envelopes are constructed by correlating current species ranges with the observed environmental conditions within those boundaries, then projecting spatially explicit climate trends into the future under different climate change scenarios to predict how species range boundaries will adjust in response (e.g. ????) (?). When predicting range shifts and species success under a changing climate, it is important to consider that range shifts driven by a single environmental variable like mean annual temperature may cause recruitment of individuals into habitats that are suboptimal in other senses, if range shifts outstrip acclimatory/adaptive potential REF. In these suboptimal habitats, range shifts may lead to reductions in local and regional species richness (Colwell et al. 2008), changes in community composition REF, ecosystem functioning (Bellard et al. 2012), and ecosystem service provision that are not predicted by the bioclimatic envelope models (Dobson et al. 2011, Isbell et al. 2011). Bioclimatic envelope models have been criticised for making a number of gross simplifications REF, INCLUDE EXAMPLES?. Basic bioclimatic envelope models assume that the breadth of the realised niche (observed spatial range) equals that of the fundamental niche (bioclimatic envelope) (Jump & Peñuelas 2005, Hoffmann & Sgrò 2011). This assumption is challenged by studies which demonstrate that the realised niche is often smaller than the fundamental niche, owing to the presence of unmeasured environmental pressures, such as biotic interactions with other species (Davis et al. 1998, Van der Putten et al. 2010, Ettinger et al. 2011). In order to accurately predict range shifts and their consequences for future biodiversity, it is important to expand bioclimatic envelope models to include variables which describe habitat as well as climatic variables such as temperature and precipitation.

For forest trees, particularly in moist tropical forests, there are often high levels of mortality during the seedling recruitment stage Coomes and Grubb 2000. Many seedlings perish due to suboptimal shade regimes created by the arrangement of adult trees REF. The seedlings of many tropical tree species are highly adapted to shade REF, meaning that if a seedling germinates in an open space, mortality by UV damage to photosynthetic machinery is quite probable. This mortality bottleneck provides a limiting factor to the success of tropical forest tree species experiencing range shifts. If seedlings germinate in areas that are only sparsely shaded, UV damage may occur leading to loss of photosynthetic capacity, reducing

49 growth rates and occasionally resulting in seedling mortality REF.

50 In montane cloud forests, range shifts are occurring more rapidly than in other areas WHY?. As mean
51 annual temperatures rise, plant species are being figuratively pushed up-slope, with higher recruitment at
52 the upslope edge of their range and higher mortality at the downslope edge of their range REF.
53 Particularly in the tropics, as altitude increases, UV-B concentration increases, with many species found at
54 high altitudes REF having specific adaptations to avoid UV-B damage to photosynthetic machinery, such
55 as WHAT?. Species found at low altitudes however, are less adapted to high UV-B environments, instead
56 having adaptations to make the most of the diminished light levels found under thick tree canopy,
57 particularly at the seedling growth stage REF. It therefore follows that as lowland species move upslope in
58 response to increasing temperature, they may experience increased levels of damaging UV-B radiation as
59 the canopy thins, but little research has been devoted to this aspect of elevational range shifts.

60 Tree species have overlapping generations, so the adult population will impact the recruitment of the next
61 generation of seedlings, through competition for water, nutrients, light and space. As species' ranges shift
62 in response to climate change, adult trees from the previous generation will still be present in the novel
63 ranges of shifting species, therefore affecting the recruitment and growth of the seedlings.

64 Need to demonstrate the knowledge gap more explicitly

65 In this study, we investigated the effects of variation in adult tree canopy structure and size distribution on
66 seedling growth form and photosynthetic stress, across an elevational gradient in the Peruvian Andes. We
67 tested two hypotheses: 1) Seedlings growing at the upslope edge of their range would experience higher
68 levels of photosynthetic stress than those at the downslope edges of their range, 2) Species would differ in
69 their acclimatory responses to changes in forest structure across the elevational gradient, resulting in
70 variation in morphological characteristics.

71 Materials and Methods

72 Study Site

73 Data collection was conducted across 10 permanent 1 ha forest plots in the Kosñipata Valley of Manú
74 National Park, Peru (-13°N, -71°W, Figure ??, Table ??). The Kosñipata Valley has been identified as a
75 migration corridor for lowland species to migrate to higher elevations in response to temperature increase
76 (?) and so is an appropriate location to study range shift drivers. Plots are situated between 400 and 3200
77 m.a.s.l. along this migration corridor (Table ??, Figure ??). The plots form part of a larger plot network
78 established by the Andes Biodiversity and Ecosystem Research Group (ABERG) in 2003 (??), and are
79 located within the "Tropical Andes" biodiversity hotspot identified in ?. The plots used in this study
80 contain 719 tree species, and the valley as a whole contains 1167 tree species ().

81 Study species

82 We chose nine tree species for comparison from a range of 1635 identified species within the 10 study plots.
83 Species were selected according to their contrasting ranges (Figure ??), differences in genus migratory
84 pattern (?), and because each species is dominant across its range in the Kosñipata Valley (ABERG,
85 unpublished data, Appendix VI). Despite having no quantitative range shift prediction information,
86 *Iriartea deltoidea* and *Dictyocaryum lamarckianum* were included in order to observe potential differences
87 in response between monocot and dicot species, as both are monocots. Both *I. deltoidea* and *D.*
88 *lamarckianum* are large-seeded palm species, as such, they are expected to be migrating upslope, similar to
89 other large-seeded palms (?). Seedlings of *Myrcia spp.* are difficult to reliably identify to species in the
90 field due to similar morphology and were thus sampled as a composite of three potential species: *Myrcia*
91 *splendens*, *M. fallax*, and *M. rostrata*, the only *Myrcia* species known to be present in our plots from
92 ABERG censuses. They are referred to as *Myrcia spp.* from here on.

93 Sampling and Measurement

94 Species were sampled in three plots representing the upper, middle and lower elevational extents of their
95 ranges (Figure ??). Within each plot, a maximum of 10 seedlings were sampled. Seedling mortality creates
96 a narrow bottleneck for tree survival in closed canopy tropical forests, seedlings are particularly sensitive
97 to environment stress. Previous studies having demonstrated seedling vulnerable to climate change (?).
98 something about dispersal not catching up at the same rate. To minimise the chance of pseudo-replication
99 of sampled seedlings, seedlings closer than 5 m to another seedling were excluded from the analysis, as it
100 cannot be guaranteed that the stems are not connected by a stolon or rhizome, it also ensures that
101 competition radius measurements are truly independent. Within a cluster of seedlings within 5 m of each
102 other, each seedling was assigned a number and a random number table was used to choose a single
103 seedling for measurement.

104 Proxies for photosynthetic efficiency or is it capacity??? were measured on the highest fully-expanded leaf
105 of each seedling. Leaf photosynthetic efficiency can be used as an indicator of physiological stress levels.
106 Plants with a lower photosynthetic efficiency are more stressed than those with a higher efficiency.
107 Chlorophyll-*a* fluorescence was measured using a Walz Mini-PAM II (Walz Effeltrich, Germany), on a
108 randomly selected area of adaxial leaf surface, avoiding prominent leaf veins according to (). Chlorophyll-*a*
109 measurements were used to calculate F_v/F_m according to ?:

$$F_v/F_m = (F_m - F_o)/F_m \quad (1)$$

110 Where F_m is the maximal fluorescence in the dark and F_o is the minimal fluorescence in the dark (?).
111 Fluorescence measurements were taken after exposing the seedling to 30 minutes of total darkness, to
112 ensure complete dark adaptation (?). Dark-adapted F_v/F_m measures the photosynthetic efficiency of the
113 leaf by relaxing the reaction centres prior to the fluorescence measurement. F_v/F_m is preferable to other
114 chlorophyll fluorescence measures as it removes the noise created by environmental conditions at the time
115 of measurement, instead providing a measure of the underlying photosynthetic efficiency. A reduction in
116 F_v/F_m is indicative of plant stress. Here, individuals with F_v/F_m values ≤ 0.7 are considered to be
117 experiencing stress (?).

118 In addition to F_v/F_m , leaf relative chlorophyll content was measured using a multi-spectral SPAD-meter
119 (Minolta SPAD-502Plus, Spectrum Technologies, Plainfield, Illinois, USA). To account for variation in
120 chlorophyll content across the leaf REF, SPAD measurements were taken at three random points on the
121 leaf. The mean of the SPAD values was used to calculate an estimate leaf chlorophyll content using the
122 conversion factor outlined in ():

$$Chl = 0.53e^{0.0364 \times SPAD} \quad (2)$$

123 Leaf and whole-plant morphological measurements

124 To assess adult-seedling competition interactions an adapted version of the Iterative Hegyi Index was
125 implemented (???). Our adapted 'Iterative Seedling Index' (ISI) uses adult tree trunk diameter at ~1.3 m
126 from ground level (Diameter at Breast Height, DBH) and the distance of trees from the seedling to
127 calculate an index for each seedling, higher values indicate greater competition pressure from surrounding
128 adult trees:

$$ISI_i = \log\left(\sum_{j=1}^n \left(\frac{1}{DIST_{ij}} D_j\right)\right) \quad (3)$$

129 where D_j is the DBH of a competitor tree and $DIST_{ij}$ is the euclidean distance between seedling i and
130 competitor tree j . ISI was log transformed for analysis, as results spanned multiple orders of magnitude.
131 The 'iterative' aspect refers to the selection of competitor trees. An iterative selection method for
132 competitive trees assumes that if the path between two trees is blocked, the intensity of competition

between them will be greatly reduced (?). The radius around the seedling is divided into 12 30° sectors, only the nearest tree >10 cm DBH within each sector is measured (Figure ??). The size of the competition radius (C_R) is defined as:

$$C_R = 2 \times \sqrt{\frac{10,000}{N}} \quad (4)$$

where N is the number of trees >10 cm DBH per ha (stand density). Stand density data was taken from ABERG census data within each plot (ABERG unpublished data) and used to interpolate values C_R for plot VC, for which no stand density data exists, by fitting the elevation of the plot to a linear regression between elevation and trees ha⁻¹ for the remaining plots (Supplementary material). C_R was rounded to the nearest metre for ease of measurement (Table ??).

Statistical Analysis

A matrix of single predictor linear mixed effects models were compared to test for the presence and strength of the causal relationship between each competition variable and each plant trait. All model variables were standardised to allow easy comparison of effect sizes, according to (???). Model quality was compared using Akaike Information Criteria (AIC) (?), Akaike weights (W_i), and fixed effect pseudo- R^2 values (R_M^2).

Add linear mixed model set up as an equation or schematic diagram or something

To better understand the potential multiplicative effects of competition variables we also compared linear mixed effects models with combinations of fixed effects, using AIC , W_i and R_M^2 , to find the model which best explained variation in each plant trait.

To understand variation between species in their physiological and morphological response to competition effects, slopes for each species were calculated and compared in the linear models.

All statistical analyses were conducted using R, version 3.2.4 (?).

Results

0.1 Variation in plant traits across elevation

Within each species, the degree to which plant traits varied over their elevational range differed.

0.2 Effects of competition on plant traits

Linear mixed effect models of the relationship between adult tree competition variables and plant traits outperformed equivalent random effect models in 15/24 cases, *i.e.* those with a $\Delta AIC_c < 2$. However, the competition variables only accounted for a small percentage of the variance in each plant trait. The highest R_M^2 in these single fixed effects models was the effect of Iterative Seedling Index on stem volume ($R_M^2 = 4.3\%$), despite the model as a whole explaining 56% of the variation in stem volume. A similar effect is seen in all the other single fixed effect models, suggesting that unmeasured site specific effects are responsible for a large portion of the variation in plant traits.

The best fitting multiple fixed effect models all included the effects of elevation, Iterative Seedling Index and Leaf Area Index to explain variation in plant traits, except leaf chlorophyll content, which only included Leaf Area Index.

168 **Discussion**

169 **Conclusion**

170 Forest structure based competition affects physiological stress independently of elevation