

Community herbivory in tropical montane rainforests is affected by phylogenetic plant diversity, specific leaf area, and leaf nutrient concentrations

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Abstract

Arthropod herbivores modulate ecosystem structure, productivity, and nutrient cycling. While previous work has shown that plant-herbivore interactions for individual species are shaped by abiotic factors, traits, and the surrounding plant community, the relative contribution of abiotic and biotic factors for herbivory at the community level remains elusive. Here, we use a structural equation model to disentangle the relative impact of temperature, precipitation (i.e., abiotic factors), tree phylogenetic diversity, and chemical as well as morphological leaf traits on total leaf herbivory in the tree canopy. Leaf area loss was estimated along an elevational gradient from 1000 to 3000 m a.s.l. in a tropical rainforest in southern Ecuador using litter traps. Community leaf herbivory ranged from 2.5 % at 3000 m to 3.4 % at 1000 m. Community leaf herbivory decreased with elevation, indirectly linked to increasing phylogenetic diversity and directly to lower specific leaf area, calcium, and iron concentrations in plants. Our results suggest that community leaf herbivory depends primarily on biotic interactions, which implies that variations in these interactions, e.g., through climate or land-use change, could cause a shift in the structure or productivity of the whole ecosystem.

Introduction

In almost all terrestrial ecosystems, herbivory caused by arthropods is an important antagonistic plant-animal interaction (Njovu *et al.* 2019). Arthropod herbivory influences ecosystems by changing the plant community composition, nutrient cycles, and plant productivity (Turcotte *et al.* 2014). Plants protect themselves using various physical and chemical defense strategies, which reduce leaf palatability (Kozlov *et al.* 2015; Zhang *et al.* 2017) and, thus, reduce leaf damage (Njovu *et al.* 2019).

Mechanical and morphometric traits, such as high leaf toughness and thickness (Coley *et al.* 1996), dry matter content (Wong *et al.* 2010), or specific leaf area (SLA; Poorter *et al.* 2004) are associated with low damage by arthropod herbivores. Further, leaf nutrient concentrations are important traits affecting herbivory (Clancy 1992; Coley *et al.* 1996; Korth *et al.* 2006; Mattson and Haack 1987; Njovu *et al.* 2019), thus leading to altered herbivore damages (Coley *et al.* 1996). For example, the ratios of carbon to nitrogen as well as nitrogen to phosphorus are inversely related to herbivore abundance (Abbas *et al.* 2014; Bergmann 2017; Njovu *et al.* 2019). Additionally, sulphur-based elicitors are known to counteract herbivory by inducing pathways of indirect plant defense (Aljbory and Chen 2018; O'Doherty *et al.* 2011) as well as activating the production of anti-feeding agents like glycosinolates (Santos *et al.* 2018). Further, foliar magnesium- (Mg), zinc- (Zn), manganese- (Mn), and iron-concentrations (Fe) generally improve herbivore fitness (Clancy 1992), although, some studies reported that these foliar components can also have toxic effects on herbivores, thus reducing leaf herbivory (Coleman *et al.* 2005; Ribeiro *et al.* 2017). Other nutrients that have been linked to arthropod herbivory include foliar potassium (K) concentration, (Heliövaara and Löyttyäniemi 1989), and aluminum (Al) concentration, which reduce herbivory if accumulated in leaves (Ribeiro *et al.* 2017).

In addition to the traits of the plant individuals, plant diversity can mediate variations in herbivory, as it influences leaf trait diversity (Hao *et al.* 2015; Vehviläinen *et al.* 2007). Phylogenetic diversity integrates information on plant traits and is thus often related to herbivory (Egorov *et al.* 2017). How the phylogenetic diversity influences herbivore communities depends on the specific composition of each community, but some reports suggest that phylogenetic diversity could be a driver for herbivory (Castagneyrol *et al.* 2014; Dinnage *et al.* 2012; Parker *et al.* 2012; Schuldt *et al.* 2014).

Besides these biotic factors, it is well-documented that climate is linked to arthropod herbivory ((Galmán *et al.* 2018; Hodkinson 2005; Kent *et al.* 2020; Kozlov *et al.* 2015; Metcalfe *et al.* 2014; Moreira *et al.* 2018; Pellissier *et al.* 2014). The most important abiotic drivers are temperature and precipitation (Bale *et al.* 2002; Jamieson *et al.* 2012). Due to their ectotherm metabolism (Hodkinson 2005), the activity and abundance of herbivorous arthropods are generally high in warm regions at low elevations (Descombes *et al.* 2017). Additionally, plant-herbivore interactions are linked to seasonal water availability (Jamieson *et al.* 2012; Janzen 1968; Kent *et al.* 2020), with higher herbivory rates during the wet season than during the dry season (Aide 1988).

As temperature, precipitation, plant traits, and phylogenetic diversity vary along elevation, analyzing herbivory patterns along elevational gradients might help to understand the specific effects of these factors. Studies looking at the interplay between abiotic- and biotic factors have observed that plants invest more energy into growth than defense under harsh climatic conditions (Coley *et al.* 1985). Moreover, herbivore pressure is lower at high elevations, thus making defense expendable (Galmán *et al.* 2018). Additionally, analyzing the variation of phylogenetic diversity across an elevational gradient and how this variation shapes leaf traits might help to understand how phylogenetic diversity affects herbivory.

In this study, we aim to disentangle the impact of abiotic factors (i.e., temperature and precipitation) and biotic factors (i.e., phylogenetic tree diversity, physical and chemical plant traits) on arthropod herbivory in three tropical montane rainforests in southern Ecuador. Over a year, we collected leaf litter of tree communities along an elevational gradient from 1000 to 3000 m a.s.l. To analyze herbivore damage, we quantified the cumulative leaf area loss across litter traps per month (Coley 1983). We hypothesize a negative correlation between increasing elevation and community leaf herbivory due to the direct and indirect effects of abiotic and biotic factors. Firstly, we expect the decrease in temperature and the increase in precipitation with increasing elevation to be negatively associated with herbivory. Secondly, we expect direct effects of temperature and precipitation on phylogenetic tree diversity. Thirdly, we hypothesize a decrease in leaf quality caused by the harsher environmental conditions at higher elevations to negatively affect leaf herbivory and lastly, we expect an indirect effect of phylogenetic tree diversity on herbivory via the leaf trait composition and thus the leaf quality.

Material & Methods

Study design

The study was conducted from February 2018 to January 2019 in the Podocarpus National Park and San Francisco reserve, in southern Ecuador (Homeier *et al.* 2008). We worked along an elevational gradient from 1000 to 3000 m a.s.l. with three main study areas: evergreen premontane forest, lower montane forest and upper montane forest (Homeier *et al.* 2008). Temperature declines along elevation, while precipitation increases (Bendix *et al.* 2008*b*; Bendix *et al.* 2008*a*). Precipitation peaks from March to August, with a dry season between September and February (Rollenbeck and Bendix 2011). At each elevation, we worked on three 100 m x 100 m plots of old-growth forest, which were located at least 100 m and at maximum 1.5 km apart from each other. Every tree with a diameter of > 10 cm at breast height within the plots was determined to the species level if possible. This data was used for the calculation of the phylogenetic diversity of each plot. Of the total species pool, we selected 52 representative tree species covering the whole variety of plant functional types within the study area. Per tree species, plant traits of 8 – 10 individuals were measured to quantify the trait variability of the tree community in each plot.

Data collection

Herbivory

To quantify leaf herbivory by arthropods at the community level of trees, we set up 18 mesh traps (1 m²) evenly distributed in the understory within each plot. The litter material within each trap was collected every two weeks. Leaf litter was scanned (Canon, CanoScan LiDE 120 with picture quality of 150 dpi) and the percentage of removed leaf area was estimated using the program WinFOLIATM (by Regent Instruments Inc., Version 2019a, Pro Version). Community

leaf herbivory was calculated for each plot as the mean percentage of leaf area loss across all measured leaves.

Plant traits

Measurements were taken from 20 mature sun-exposed leaves without herbivore damage from each tree (for four species only 5 – 7 individuals were found and for a few species with particularly big leaves, a smaller number of leaves was analyzed due to time constraints). We estimated foliar C, P, Al, Ca, Fe, K, Mg, Mn, S, and N concentrations, CN ratio, and NP ratio, as well as specific leaf area (SLA), leaf area (LA), leaf thickness, leaf toughness, and dry matter content (DMC). For the quantification of leaf thickness and leaf toughness, three additional leaves per tree individual were used. For details see Homeier *et al.* (2021).

Trait measurements were taken from 422 trees belonging to 52 species (Tab. A1), with several missing values for single traits (Tab. A2). Missing values were replaced with mean values of the respective plot. To be able to make inferences on a community level, we calculated community-weighted means (CWM) for each plant trait. This was done using the R package *stats* (R Core Team 2021) by first computing mean trait values as well as abundance values for each species per plot and then using the *weighted.mean* function to get CWM trait values per plot.

Phylogenetic diversity

A phylogenetic tree was built with the online tool Phylomatic (<https://phylodiversity.net/phyloomatic/>; Webb and Donoghued 2005), based on the backbone mega tree S03_R20160415_euphylllophyte.new provided by M. Gastauer and J. Meira-Neto. Age calibration was done using the method suggested by Grafen (1989) implemented in the package *ape* (Paradis and Schliep 2019). We calculated mean pairwise phylogenetic distance (MPD) per plot based on the previously constructed phylogenetic tree using the *ses.mpd* function of the R package *picante* (Kembel *et al.* 2010). Hereby, the function parameter *null.model* was set to *independent swapping* and abundance weighting was activated.

Climate data

Temperature and precipitation were measured using weather stations within each plot. From January 2018 to December 2020, we sampled daily precipitation as well as daily mean temperature. Using these data, we calculated the total precipitation and mean temperature for each month and plot.

Statistical Analysis

The statistical analysis was performed using R Version 4.0.3 (R Core Team 2020). All leaf trait and climate data were z-transformed and, additionally, leaf trait data were log₁₀-transformed. We tested for collinearity between plant traits using the *Hmisc* package (Harell, Jr. and Dupont 2020). According to these collinearity tests and literature research regarding their expected impact on herbivory, we selected the following CWM plant traits for further analyses: SLA, S,

Ca, Fe, Mn, CN, and NP (Appendix Fig. A1, Tab. A3). We used backward stepwise selections to identify the minimal adequate linear mixed-effects models predicting community leaf herbivory from leaf traits, leaf traits from phylogenetic diversity, and phylogenetic diversity from abiotic data by using the R package *lmerTest* (Kuznetsova *et al.* 2020). Month and elevational levels were included as random effects.

We used structural equation modeling (SEM) to estimate direct and indirect effects of abiotic (temperature and precipitation) and biotic (leaf traits as well as MPD between trees) parameters on community leaf herbivory, using the R package *piecewiseSEM* (Lefcheck 2016). Here, we used monthly means per plot for community leaf herbivory and abiotic data to incorporate temporal variations into our model. We time-shifted temperature by 3 months, as the correlation between community leaf herbivory and temperature peaked for this time-shift (Tab. A4). The elevational level as well as the month of sampling were included as random effects. Additionally, we looked for seasonal patterns in herbivory at the different elevational levels. Therefore, we plotted monthly means of community leaf herbivory per elevational level against time to visualize how herbivory varied over the course of the year.

Results

On average, we collected 2033, 2406, and 2512 leaves per plot at 1000, 2000, and 3000 m a.s.l., respectively. Herbivory and CWM Ca concentration exhibited a humped-shaped curve across elevation. CWM SLA continually decreased across the elevational gradient. CWM Fe concentration decreased from 1000 to 2000 m a.s.l., but then stayed the same at 3000 m a.s.l. MPD continually increased with increasing elevation (Tab. A5) changing from overdispersion to clustering.

Linear model backward stepwise selection identified CWM SLA, Fe-, and Ca concentrations as the best predictors for community leaf herbivory (Tab A6). SEM showed that community leaf herbivory was impacted positively by SLA, directly negatively by Ca and directly negatively by Fe concentrations (Fig. 1; Fig. 2). Herbivory was impacted indirectly by MPD (Fig. 1; Fig. 2). There was no significant impact of temperature nor precipitation on any of the response variables (Fig. 1). There was considerable variation of community leaf herbivory across the year at all three elevational levels, ranging from 2.2 % in July to 7.0 % in April (Tab. A7). Monthly variation was the most pronounced at 1000 m a.s.l. ranging from 2.1 % in January to 9.6 % in April (Fig. 3).

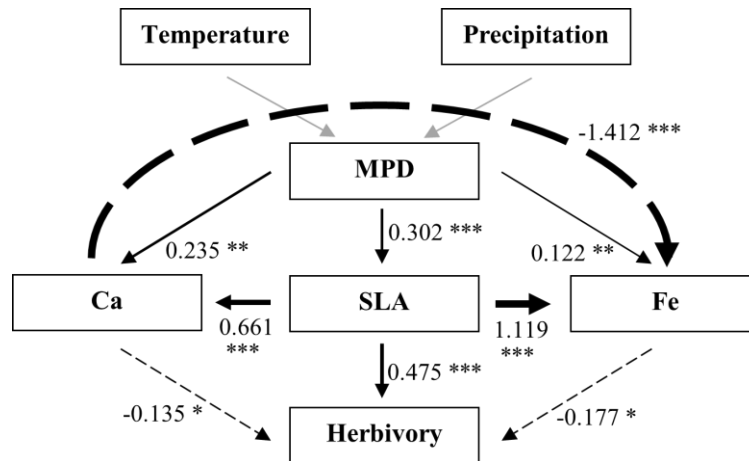


Fig. 1: Associations of abiotic and biotic factors with community leaf herbivory in a south Ecuadorian montane rainforest depicted as structural equation model linking mean monthly temperature [$^{\circ}\text{C}$], mean monthly precipitation [mm] with mean pairwise phylogenetic distance (MPD) with community weighted mean SLA [cm^2/g], Ca- [mg/g DM], and Fe [mg/g DM] concentration, and mean monthly community leaf herbivory (percentual leaf area loss [cm^2]) along an elevational gradient from 1000 to 3000 m a.s.l. The value of the estimated effect size is depicted in the thickness of the arrows. Black and solid arrows indicate significant positive effects, dashed arrows indicate significant negative effects, grey arrows indicate non significance. Asterisks mark the significance level ($0.050 > * < 0.010 > ** < 0.001 > *** < 0.000$).

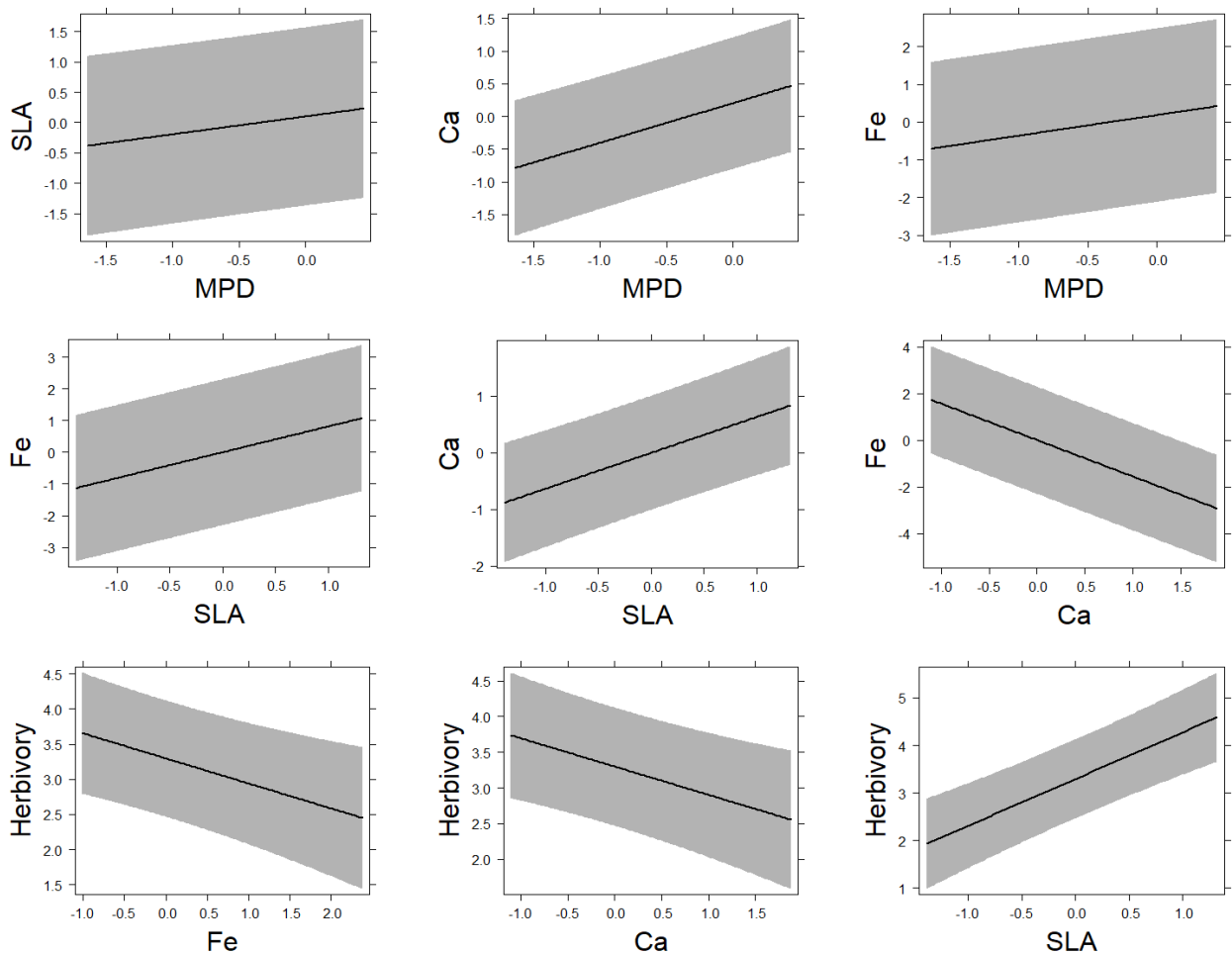


Fig. 2: Significant relationships between abiotic and biotic factors of the structural equation

model analyzing the influence of temperature [°C], precipitation [mm], mean pairwise distance (MPD), and community weighted mean calcium (Ca) [mg/g DM], iron (Fe) [mg/g DM], and specific leaf area (SLA) [cm²/g] on community leaf herbivory [%] with regression line (black) and 95% confidence interval (grey) in a south Ecuadorian tropical montane rainforest along an elevational gradient from 1000 to 3000 m a.s.l.

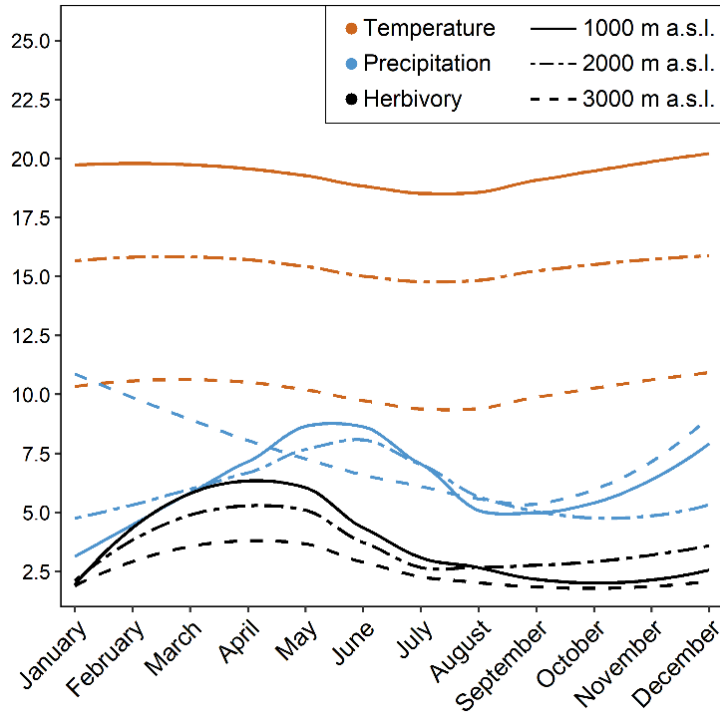


Fig. 3: Mean monthly community leaf herbivory [%], temperature [°C], and mean daily precipitation [mm] in a south Ecuadorian tropical montane rainforest from 1000 to 3000 m a.s.l. over one year.

Discussion

Our study on community leaf herbivory along an elevational gradient revealed that community leaf herbivory was directly and positively associated with the community weighted mean (CWM) leaf trait SLA, and negatively to the CWM Ca-, and Fe concentrations. MPD was directly positively associated with all traits and thus indirectly associated with community leaf herbivory. Unexpectedly, neither temperature nor precipitation were related to community herbivory.

The observed positive effect of SLA on community leaf herbivory matches past studies (Garibaldi *et al.* 2011; Poorter *et al.* 2004; Schädler *et al.* 2003). SLA was already used as a predictor for herbivory (Bendix *et al.* 2021; Hulshof *et al.* 2013) since low SLA indicates reduced palatability due to high leaf thickness and low leaf water content (Coley *et al.* 1996; Marengo *et al.* 2009). Additionally, our results support the herbivore defensive function of foliar Ca- and Fe concentrations. The negative relation between Ca concentrations and herbivory can be a result of chemical defense against herbivores as plants use Ca to build Ca oxalate minerals, which were found in most terrestrial plants (Hanley *et al.* 2007) and have a detrimental effect on chewing insects (Korth *et al.* 2006). Similarly, Fe concentrations, which are negatively

related to community leaf herbivory in our study have been shown to reduce leaf herbivory (Ribeiro *et al.* 2017).

As expected, community leaf herbivory was indirectly impacted by MPD through leaf nutritional quality. We observed an increase in tree phylogenetic diversity with increasing elevation, but contrary to the literature (McCain and Colwell 2011; Sarano 2019) we could not translate this trend to the decreasing temperature along our elevational gradient. This change in tree phylogenetic diversity was related to the three leaf traits directly associated with the community leaf herbivory, either with a positive relation (SLA) or a negative relation (foliar Ca and Fe concentrations). We did not find a direct association between tree phylogenetic diversity and herbivory. Since both herbivorous- and predatory arthropod diversity as well as abundance benefit from increased plant phylogenetic diversity, its potential association with community leaf herbivory could be outweighed by the herbivores' predators (Dinnage *et al.* 2012).

The positive relation of phylogenetic diversity with CWM SLA resulted in a well-documented decline of SLA along elevation (Hulshof *et al.* 2013). This could be explained by evolutionary adaptations of tree species to harsher environmental conditions, like poor resource availability, which shape the SLA (Ackerly 2004). Leaves with smaller SLAs are better adapted to resource-poor environments because they allow for more resource retention due to their longer life span (Wilson *et al.* 1999). Resource availability generally decreases with elevation, and it has been demonstrated that there is poor nutrient availability at high elevations in our study area, as soil acidity, CN ratio, and soil moisture are elevated (Moser *et al.* 2011). In accordance with the existing literature, we found a positive relationship between phylogenetic diversity with foliar concentrations of Ca and Fe (Hao *et al.* (2015), and a direct negative association between Ca and Fe concentrations (Gunes *et al.* 1998) supporting the notion that food quality of leaves being shaped by nutrient concentration as well as elemental defense (Boyd 2007). Moreover, the observed positive association of SLA on concentrations of both Ca and Fe reflects a robust relationship previously documented across biomes (Hoffmann *et al.* 2005). These relationships between leaf traits could have been promoted by the strong correlations between many measured leaf traits in our study, indicating complex foliar nutrient interactions. It should be noted that both, MPD and the CWM leaf traits were not measured from the same leaves or tree species, from which leaf herbivory was determined. However, as we took care to take representative samples from the tree community, we feel that our study allows in general reliable conclusions.

Although a multitude of previous studies have found that temperature plays a key role in leaf herbivory (Bale *et al.* 2002; Cornelissen 2011; Janzen 1968; Loughnan and Williams 2019), we observed no such effect. The relation between temperature and leaf herbivory can mostly be traced back to correlated patterns in arthropod abundance or activity as temperature influences the metabolic activity of arthropods (Aide 1988; Aide 1993; Lemoine *et al.* 2014). As such, reduced temperatures at high elevations potentially lead to an overall decrease in herbivore pressure (Descombes *et al.* 2017). We suspect the absence of this effect is because temperatures in our study area remained sufficiently high to support insect activity across all elevations.

We also did not find an effect of precipitation on community leaf herbivory, although several studies found either a negative or positive precipitation effect across different climatic regions (Janzen 1968; Kent *et al.* 2020; Kozlov *et al.* 2015; Loughnan and Williams 2019; Mazía *et al.* 2012; Rodríguez-Castañeda 2013; Vasconcelos 1999). However, studies demonstrating precipitation effects on herbivory typically investigated water-stressed plants (Mattson and Haack 1987). Due to overall high water availability along the elevational gradient in our study area, plants do not suffer water stress (Rollenbeck and Bendix 2011), suggesting that variations in precipitation are not strong enough to cause significant effects on herbivory.

Precipitation has also often been linked to herbivory via seasonality (Mazía *et al.* 2012). Hereby, studies observed higher herbivory rates during wetter months in response to elevated insect abundance (Aide 1988; Aide 1993). This increase in abundance might be due to the great production of young leaves with high palatability as a consequence of the increased water availability (Coley 1983; Wolda 1978). We also found a clear seasonal pattern in community herbivory across the elevational gradient, with a peak in April. Here it becomes apparent, that because we did not find evidence that precipitation is the driver behind this seasonal variation in herbivory, the temporal development of the herbivorous arthropods should be investigated further in future research.

Conclusion

We found a complex interplay of community leaf herbivory, phylogenetic plant diversity, and leaf traits, which resulted in decreased community leaf herbivory at higher elevations. We found a direct and positive effect of increasing phylogenetic plant diversity on all leaf traits. Further, we demonstrated a positive link between the leaf trait SLA and community leaf herbivory and negative relations of Ca-, and Fe concentrations on community leaf herbivory. Furthermore, we present evidence that contrary to the literature the abiotic factors temperature and precipitation are not necessarily drivers for variation in community leaf herbivory in tree canopies of tropical montane rainforests. We detected clear seasonal variations in community leaf herbivory with higher leaf area loss during wetter months than dryer months. For future research, we recommend repeated measurements over one year to gain a better understanding of the direct and indirect interactions between abiotic and biotic factors and community leaf herbivory. This will help to gain a mechanistic understanding of how varying abiotic factors modulate leaf herbivory on a community level and thus influence ecosystem functions such as primary productivity.

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References

- Abbas, M., Klein, A.-M., Ebeling, A., Oelmann, Y., Ptacnik, R., Weisser, W. W. and Hillebrand, H. (2014): Plant diversity effects on pollinating and herbivorous insects can be linked to plant stoichiometry. – *Basic and Applied Ecology* **15**(2): 169–178. <http://dx.doi.org/10.1016/j.baae.2014.02.001>
- Ackerly, D. D. (2004): Adaptation, niche conservatism, and convergence: comparative studies of leaf evolution in the California chaparral. – *The American naturalist* **163**(5): 654–671. <http://dx.doi.org/10.1086/383062>
- Aide, M. T. (1993): Patterns of leaf development and herbivory in a tropical understory community. – *Ecology* **74**(2): 455–466. <http://dx.doi.org/10.2307/1939307>
- Aide, T. M. (1988): Herbivory as selective agent on the timing of leaf production in a tropical understory community. – *Nature* **336**: 574–575. <http://dx.doi.org/10.1038/336574a0>
- Aljbory, Z. and Chen, M.-S. (2018): Indirect plant defense against insect herbivores: a review. – *Insect science* **25**(1): 2–23. <http://dx.doi.org/10.1111/1744-7917.12436>
- Bale, J. S., Masters, G. J., Hodkinson, I. D., Awmack, C., Bezemer, Brown, V. K., Butterfield, J., Buse, A., Coulson, J. C., Farrar, J., Good, J. E. G., Harrington, R., Hartley, S., Jones, T. H., Lindroth, R. L., Press, M. C., Symrnioudis, I., Watt, A. D. and Whittaker, J. B. (2002): Herbivory in global climate change research: direct effects of rising temperature on insect herbivores. – *Global Change Biology* **8**: 1–16. <http://dx.doi.org/10.1046/j.1365-2486.2002.00451.x>
- Bendix, J., Aguire, N., Beck, E., Bräuning, A., Brandl, R., Breuer, L., Böhning-Gaese, K., Paula, M. D. de, Hickler, T., Homeier, J., Inclan, D., Leuschner, C., Neuschulz, E. L., Schleuning, M., Suarez, J. P., Trachte, K., Wilcke, W., Windhorst, D. and Farwig, N. (2021): A research framework for projecting ecosystem change in highly diverse tropical mountain ecosystems. – *Oecologia* **195**(3): 589–600. <http://dx.doi.org/10.1007/s00442-021-04852-8>
- Bendix, J., Rollenbeck, R., Fabian, P., Emck, P., Richter, M. and Beck, E. (2008a): *Climate Variability*. – In: Beck, E., Bendix, J., Kottke, I., Makeschin, F. and Mosandl, R. (eds): *Gradients in a Tropical Mountain Ecosystem of Ecuador*. Ecological Studies, No. 198. Springer, Berlin, Heidelberg, pp. 281–290.
- Bendix, J., Rollenbeck, R., Richter, M., Fabian, P. and Emck, P. (2008b): *Climate*. – In: Beck, E., Bendix, J., Kottke, I., Makeschin, F. and Mosandl, R. (eds): *Gradients in a Tropical*

- Mountain Ecosystem of Ecuador. Ecological Studies, No. 198. Springer, Berlin, Heidelberg, pp. 63–73.
- Bergmann, A. (2017): *Variation of herbivory after nitrogen and/or phosphor addition to Ecuadorian tropical montane forests*, Master's thesis, Goettingen.
- Boyd, R. S. (2007). The defense hypothesis of elemental hyperaccumulation: status, challenges and new directions. *Plant and Soil*, 293, 153–176.
- Castagneyrol, B., Jactel, H., Vacher, C., Brockerhoff, E. G. and Koricheva, J. (2014): Effects of plant phylogenetic diversity on herbivory depend on herbivore specialization. – *J Appl Ecol* **51**(1): 134–141. <http://dx.doi.org/10.1111/1365-2664.12175>
- Clancy, K. M. (1992): Response of western spruce budworm (Lepidoptera: Tortricidae) to increased nitrogen in artificial diets. – *Environmental Entomology* **21**(2): 331–344. <http://dx.doi.org/10.1093/ee/21.2.331>
- Coleman, C. M., Boyd, R. S. and Eubanks, M. D. (2005): Extending the elemental defense hypothesis: dietary metal concentrations below hyperaccumulator levels could harm herbivores. – *Journal of chemical ecology* **31**(8): 1669–1681. <http://dx.doi.org/10.1007/s10886-005-5919-4>
- Coley, P. D. (1983): Herbivory and defense characteristics of tree species in a lowland tropical forest. – *Ecological Monographs* **53**(2): 209–234. <http://dx.doi.org/10.2307/1942495>
- Coley, P. D., Barone and J. A. (1996): Herbivory and plant defenses in tropical forests. – *Annual Review of Ecology, and Systematics* **27**: 305–335. <http://dx.doi.org/10.1146/annurev.ecolsys.27.1.305>
- Coley, P. D., Byrant, J. P. and Chapin, F. S. (1985): Resource availability and plant antiherbivore defense. – *Science* **230**(4728): 895–899. <http://dx.doi.org/10.1126/science.230.4728.895>
- Cornelissen, T. (2011): Climate change and its effects on terrestrial insects and herbivory patterns. – *Neotropical Entomology* **40**(2): 155–163. <http://dx.doi.org/10.1590/S1519-566X2011000200001>
- Descombes, P., Marchon, J., Pradervand, J.-N., Bilat, J., Guisan, A., Rasmann, S. and Pellissier, L. (2017): Community-level plant palatability increases with elevation as insect herbivore abundance declines. – *Journal of Ecology* **105**(1): 142–151. <http://dx.doi.org/10.1111/1365-2745.12664>
- Dinnage, R., Cadotte, M. W., Haddad, N. M., Crutsinger, G. M. and Tilman, D. (2012): Diversity of plant evolutionary lineages promotes arthropod diversity. – *Ecology letters* **15**(11): 1308–1317. <http://dx.doi.org/10.1111/j.1461-0248.2012.01854.x>
- Egorov, E., Gossner, M. M., Meyer, S. T., Weisser, W. W. and Brändle, M. (2017): Does plant phylogenetic diversity increase invertebrate herbivory in managed grasslands? – *Basic and Applied Ecology* **20**: 40–50. <http://dx.doi.org/10.1016/j.baae.2017.03.004>
- Galmán, A., Abdala-Roberts, L., Zhang, S., Berny-Mier y Teran, J. C., Rasmann, S. and Moreira, X. (2018): A global analysis of elevational gradients in leaf herbivory and its

- underlying drivers: Effects of plant growth form, leaf habit and climatic correlates. – *Journal of Ecology* **106**(1): 413–421. <http://dx.doi.org/10.1111/1365-2745.12866>
- Garibaldi, L. A., Kitzberger, T. and Chaneton, E. J. (2011): Environmental and genetic control of insect abundance and herbivory along a forest elevational gradient. – *Oecologia* **167**(1): 117–129. <http://dx.doi.org/10.1007/s00442-011-1978-0>
- Grafen, A. (1989): The Phylogenetic Regression. – *Biological Sciences* **326**(1233): 119–157. <http://dx.doi.org/10.1098/rstb.1989.0106>
- Gunes, A., Alpaslan, M. and Inal, A. (1998): Critical nutrient concentrations and antagonistic and synergistic relationships among the nutrients of NFT-grown young tomato plants. – *Journal of Plant Nutrition* **21**(10): 2035–2047. <http://dx.doi.org/10.1080/01904169809365542>
- Hanley, M. E., Lamont, B. B., Fairbanks, M. M. and Rafferty, C. M. (2007): Plant structural traits and their role in anti-herbivore defence. – *Perspectives in Plant Ecology, Evolution and Systematics* **8**(4): 157–178. <http://dx.doi.org/10.1016/j.ppees.2007.01.001>
- Hao, Z., Kuang, Y. and Kang, M. (2015): Untangling the influence of phylogeny, soil and climate on leaf element concentrations in a biodiversity hotspot. – *Funct Ecol* **29**(2): 165–176. <http://dx.doi.org/10.1111/1365-2435.12344>
- Harell, F. E., Jr. and Dupont, C. (2020): *Hmisc: Harell Miscellaneous*: R package version.
- Heliövaara, K. and Löyttyniemi, K. (1989): Effect of forest fertilization on pine needle-feeding Coleoptera. – *Silva Fennica* **23**(4): 279–286.
- Hodkinson, I. D. (2005): Terrestrial insects along elevation gradients: Species and community responses to altitude. – *Biological reviews of the Cambridge Philosophical Society* **80**(3): 489–513. <http://dx.doi.org/10.1017/s1464793105006767>
- Hoffmann, W. A., Franco, A. C., Moreira, M. Z. and Haridasan, M. (2005): Specific leaf area explains differences in leaf traits between congeneric savanna and forest trees. – *Funct Ecology* **19**(6): 932–940. <http://dx.doi.org/10.1111/j.1365-2435.2005.01045.x>
- Homeier, J., Seeler, T., Pierick, K. and Leuschner, C. (2021): Leaf trait variation in species-rich tropical Andean forests. – *Scientific reports* **11**(1): 9993. <http://dx.doi.org/10.1038/s41598-021-89190-8>
- Homeier, J., Werner, F. A., Gradstein, S. R., Breckle, S. W. and Richter, M. (2008): *Flora and Fungi: Composition and function: Potential vegetation and floristic composition of Andean forests in South Ecuador*. – In: Beck, E., Bendix, J., Kottke, I., Makeschin, F. and Mosandl, R. (eds): *Gradients in a Tropical Mountain Ecosystem of Ecuador*. Ecological Studies, No. 198. Springer, Berlin, Heidelberg, pp. 87–100.
- Hulshof, C. M., Violle, C., Spasojevic, M. J., McGill, B., Damschen, E., Harrison, S. and Enquist, B. J. (2013): Intra-specific and inter-specific variation in specific leaf area reveal the importance of abiotic and biotic drivers of species diversity across elevation and latitude. – *Journal of Vegetation Science* **24**(5): 921–931. <http://dx.doi.org/10.1111/jvs.12041>
- Jamieson, M. A., Trowbridge, A. M., Raffa, K. F. and Lindroth, R. L. (2012): Consequences of climate warming and altered precipitation patterns for plant-insect and multitrophic

- interactions. – *Plant physiology* **160**(4): 1719–1727.
<http://dx.doi.org/10.1104/pp.112.206524>
- Janzen, D. H. (1968): Differences in Insect Abundance and Diversity Between Wetter and Drier Sites During a Tropical Dry Season. – *Ecology* **49**(1): 96–110.
<http://dx.doi.org/10.2307/1933565>
- Kembel, S. W., Ackerly, D. D., Blomberg, S. P., Cornwell, W. K., Cowan, P. D., Helmus, M. R., Morlon, H. and Webb, C. O. (2010): Picante: Integrating Phylogenies and Ecology. – *Bioinformatics* **26**(11): 1463–1464. <http://dx.doi.org/10.1093/bioinformatics/btq166>
- Kent, D. R., Lynn, J. S., Pennings, S. C., Souza, L. A., Smith, M. D. and Rudgers, J. A. (2020): Weak latitudinal gradients in insect herbivory for dominant rangeland grasses of North America. – *Ecology and Evolution* **10**(13): 6385–6394.
<http://dx.doi.org/10.1002/ece3.6374>
- Korth, K. L., Doege, S. J., Park, S.-H., Goggin, F. L., Wang, Q., Gomez, S. K., Liu, G., Jia, L. and Nakata, P. A. (2006): Medicago truncatula mutants demonstrate the role of plant calcium oxalate crystals as an effective defense against chewing insects. – *Plant physiology* **141**(1): 188–195. <http://dx.doi.org/10.1104/pp.106.076737>
- Kozlov, M. V., Lanta, V., Zverev, V. and Zvereva, E. L. (2015): Global patterns in background losses of woody plant foliage to insects. – *Global Ecology and Biogeography* **24**(10): 1126–1135. <http://dx.doi.org/10.1111/geb.12347>
- Kuznetsova, A., Brockhoff, P. B., Christensen, R. H. B. and Jensen, S. P. (2020): *lmerTest*: Tests in Linear Mixed Effects Models.
- Lefcheck, J. S. (2016): piecewiseSEM : Piecewise structural equation modelling in R for ecology, evolution, and systematics. – *Methods in Ecology and Evolution* **7**(5): 573–579.
<http://dx.doi.org/10.1111/2041-210X.12512>
- Lemoine, N. P., Bukepile, D. E. and Parker, J. D. (2014): *Variable effects of temperature on insect herbivory*. <https://doi.org/10.7717/peerj.376>. Accessed March 10, 2021.
- Limberger, O., Homeier, J., Farwig, N., Pucha-Cofrep, F., Fries, A., Leuschner, C., Trachte, K. and Bendix, J. (2021): Classification of Tree Functional Types in a Megadiverse Tropical Mountain Forest from Leaf Optical Metrics and Functional Traits for Two Related Ecosystem Functions. – *Forests* **12**(5): 649. <http://dx.doi.org/10.3390/f12050649>
- Loughnan, D. and Williams, J. L. (2019): Climate and leaf traits, not latitude, explain variation in plant-herbivore interactions across a species' range. – *Journal of Ecology* **107**(2): 913–922. <http://dx.doi.org/10.1111/1365-2745.13065>
- Marenco, R. A., Antezana-Vera, S. A. and Nascimento, H.C.S. (2009): Relationship between specific leaf area, leaf thickness, leaf water content and SPAD-502 readings in six Amazonian tree species. – *Photosynthetica* **47**(2): 184–190.
<http://dx.doi.org/10.1007/s11099-009-0031-6>
- Mattson, W. J. and Haack, R. A. (1987): The role of drought in outbreaks of plant-eating insects. – *BioSciences* **37**(2): 110–118. <http://dx.doi.org/10.2307/1310365>
- Mazía, N., Chaneton, E. J., Dellacanonica, C., Dipaolo, L. and Kitzberger, T. (2012): Seasonal patterns of herbivory, leaf traits and productivity consumption in dry and wet

- Patagonian forests. – *Ecological Entomology* **37**(3): 193–203.
<http://dx.doi.org/10.1111/j.1365-2311.2012.01356.x>
- McCain, C. M. and Colwell, R. K. (2011): Assessing the threat to montane biodiversity from discordant shifts in temperature and precipitation in a changing climate. – *Ecology letters* **14**(12): 1236–1245. <http://dx.doi.org/10.1111/j.1461-0248.2011.01695.x>
- Metcalfe, D. B., Asner, G. P., Martin, R. E., Silva Espejo, J. E., Huasco, W. H., Farfán Amézquita, F. F., Carranza-Jimenez, L., Galiano Cabrera, D. F., Baca, L. D., Sinca, F., Huaraca Quispe, L. P., Taype, I. A., Mora, L. E., Dávila, A. R., Solórzano, M. M., Puma Vilca, B. L., Laupa Román, J. M., Guerra Bustios, P. C., Revilla, N. S., Tupayachi, R., Girardin, C. A. J., Doughty, C. E. and Malhi, Y. (2014): Herbivory makes major contributions to ecosystem carbon and nutrient cycling in tropical forests. – *Ecology letters* **17**(3): 324–332. <http://dx.doi.org/10.1111/ele.12233>
- Moreira, X., Castagneyrol, B., Abdala-Roberts, L., Berny-Mier y Teran, J. C., Timmermans, B. G. H., Bruun, H. H., Covelo, F., Glauser, G., Rasmann, S. and Tack, A. J. M. (2018): Latitudinal variation in plant chemical defences drives latitudinal patterns of leaf herbivory. – *Ecography* **41**(7): 1124–1134. <http://dx.doi.org/10.1111/ecog.03326>
- Moser, G., Leuschner, C., Hertel, D., Graefe, S., Soethe, N. and Iost, S. (2011): Elevation effects on the carbon budget of tropical mountain forests (S Ecuador): the role of the belowground compartment. – *Global Change Biology* **17**(6): 2211–2226.
<http://dx.doi.org/10.1111/j.1365-2486.2010.02367.x>
- Njovu, H. K., Peters, M. K., Schellenberger Costa, D., Brandl, R., Kleyer, M. and Steffan-Dewenter, I. (2019): Leaf traits mediate changes in invertebrate herbivory along broad environmental gradients on Mt. Kilimanjaro, Tanzania. – *The Journal of animal ecology* **88**(11): 1777–1788. <http://dx.doi.org/10.1111/1365-2656.13058>
- O'Doherty, I., Yim, J. J., Schmelz, E. A. and Schroeder, F. C. (2011): Synthesis of caeliferins, elicitors of plant immune responses: accessing lipophilic natural products via cross metathesis. – *Organic letters* **13**(21): 5900–5903. <http://dx.doi.org/10.1021/ol202541b>
- Paradis, E. and Schliep, K. (2019): ape 5.0: an environment for modern phylogenetics and evolutionary analyses in R. – *Bioinformatics (Oxford, England)* **35**(3): 526–528.
<http://dx.doi.org/10.1093/bioinformatics/bty633>
- Parker, J. D., Burkepille, D. E., Lajeunesse, M. J. and Lind, E. M. (2012): Phylogenetic isolation increases plant success despite increasing susceptibility to generalist herbivores. – *Diversity and Distributions* **18**(1): 1–9. <http://dx.doi.org/10.1111/j.1472-4642.2011.00806.x>
- Pellissier, L., Roger, A., Bilat, J. and Rasmann, S. (2014): High elevation *Plantago lanceolata* plants are less resistant to herbivory than their low elevation conspecifics: is it just temperature? – *Ecography* **37**(10): 950–959. <http://dx.doi.org/10.1111/ecog.00833>
- Poorter, L., van de Plassche, M., Willems, S. and Boot, R. G. A. (2004): Leaf traits and herbivory rates of tropical tree species differing in successional status. – *Plant Biology* **6**(6): 746–754. <http://dx.doi.org/10.1055/s-2004-821269>

- R Core Team (2021): *R: A language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna, Austria.
- Ribeiro, S. P., Londe, V., Bueno, A. P., Barbosa, J. S., Corrêa, T. L., Soeltl, T., Maia, M., Pinto, V. D., França Dueli, G. de, Sousa, H. C. de, Kozovits, A. R. and Nalini, H. A. (2017): Plant defense against leaf herbivory based on metal accumulation: examples from a tropical high altitude ecosystem. – *Plant Species Biol.* **32**(2): 147–155. <http://dx.doi.org/10.1111/1442-1984.12136>
- Rodríguez-Castañeda, G. (2013): The world and its shades of green: A meta-analysis on trophic cascades across temperature and precipitation gradients. – *Global Ecology and Biogeography* **22**(1): 118–130. <http://dx.doi.org/10.1111/j.1466-8238.2012.00795.x>
- Rollenbeck, R. and Bendix, J. (2011): Rainfall distribution in the Andes of southern Ecuador derived from blending weather radar data and meteorological field observations. – *Atmospheric Research* **99**(2): 277–289. <http://dx.doi.org/10.1016/j.atmosres.2010.10.018>
- Santos, N. A., Teixeira, N. C., Valim, J. O. S., Almeida, E. F. A., Oliveira, M. G. A. and Campos, W. G. (2018): Sulfur fertilization increases defense metabolites and nitrogen but decreases plant resistance against a host-specific insect. – *Bulletin of entomological research* **108**(4): 479–486. <http://dx.doi.org/10.1017/S0007485317001018>
- Sarano, F. R. (2019): *Biodiversity sector: risks of temperature increase to biodiversity and ecosystems*. – In: Nobre, C., Marengo, J. and Soares W. (eds): *Climate change risks in Brazil*. Springer, Cham.
- Schädler, M., Jung, G., Auge, H. and Brandl, R. (2003): Palatability, decomposition and insect herbivory: Patterns in a successional old-field plant community. – *OIKOS* **103**: 121–132. <http://dx.doi.org/10.1034/j.1600-0706.2003.12659.x>
- Schuldt, A., Assmann, T., Bruelheide, H., Durka, W., Eichenberg, D., Härdtle, W., Kröber, W., Michalski, S. G. and Purschke, O. (2014): Functional and phylogenetic diversity of woody plants drive herbivory in a highly diverse forest. – *The New phytologist* **202**(3): 864–873. <http://dx.doi.org/10.1111/nph.12695>
- Turcotte, M. M., Thomsen, C. J. M., Broadhead, G. t., Fine, P. V. A., Godfrey, R. M., Lamarre, G. P.A., Meyer, S. T., Richards, L. A. and Johnson, M. T. J. (2014): Percentage leaf herbivory across vascular plant species. – *Ecology* **95**(3): 788. <http://dx.doi.org/10.1890/13-1741.1>
- Vasconcelos, H. L. (1999): Levels of leaf herbivory in Amazonian trees from different stages in forest regeneration. – *Acta Amazonica* **29**(4): 615–623. <http://dx.doi.org/10.1590/1809-43921999294623>
- Vehviläinen, H., Koricheva, J. and Ruohomäki, K. (2007): Tree species diversity influences herbivore abundance and damage: meta-analysis of long-term forest experiments. – *Oecologia* **152**(2): 287–298. <http://dx.doi.org/10.1007/s00442-007-0673-7>
- Webb, C. O. and Donoghued, M. J. (2005): Phylomatic: tree assembly for applied phylogenetics. – *Mol Ecol Notes* **5**(1): 181–183. <http://dx.doi.org/10.1111/j.1471-8286.2004.00829.x>

- Whittaker, R. H. (2012): *The biochemical ecology of higher plants*. – In: Sondheimer, E. (ed.): *Chemical Ecology*. Academic Press, London, pp. 43–102.
- Wilson, P. J., Thompson, K. and Hodgson, J. G. (1999): Specific leaf area and leaf dry matter content as alternative predictors of plant strategies. – *New Phytologist* **143**: 155–162. <http://dx.doi.org/10.1046/j.1469-8137.1999.00427.x>
- Wolda, H. (1978): Seasonal fluctuations in rainfall, food and abundance of tropical insects. – *Journal of Animal Ecology* **47**(2): 369–381. <http://dx.doi.org/10.2307/3789>
- Wong, P. K.K., Liang, Y. A.N., Liu, N. Y.A. and Qiu, J.-W. (2010): Palatability of macrophytes to the invasive freshwater snail *Pomacea canaliculata*: differential effects of multiple plant traits. – *Freshwater Biology* **55**(10): 2023–2031. <http://dx.doi.org/10.1111/j.1365-2427.2010.02458.x>
- Zhang, S., Zhang, Y. and Ma, K. (2017): The association of leaf lifespan and background insect herbivory at the interspecific level. – *Ecology* **98**(2): 425–432. <http://dx.doi.org/10.1002/ecy.1649>

Appendix

Tab. A1: Number of samples per elevational level. Measurements were taken from the indicated number of individual trees belonging to the indicated number of species.

Elevation [m a.s.l.]	Plant traits		Phylogenetic diversity	
	species	trees	species	trees
1000	20	156	239	1249
2000	20	169	384	3386
3000	12	97	217	3877
Total	52	422	509	8512

Tab. A2: Number of sampled tree individuals with measured and missing values per leaf trait.

	C	P	Al	Ca	Fe	K	Mg	Mn	S	N	CN	NP	SLA	LA	Thick- ness	Tough- ness	DMC
Measured	416	418	405	418	418	393	418	418	376	416	416	414	420	378	421	422	421
Missing	6	4	17	4	4	29	4	4	46	6	6	8	2	44	1	0	1

Table A3: Selection criteria of plant traits to be used in the community herbivory analyses.

Trait	Notes	References
Specific Leaf Area (SLA) [cm ² /g]	Correlation with palatability, therefore, could affect herbivory; Association to physical leaf defense	Schädler <i>et al.</i> (2003) Descombes <i>et al.</i> (2017)
Sulphur (S) [mg/g DM]	Component of some elicitors and anti-feeding agents	Aljbery and Chen (2018) O'Doherty <i>et al.</i> (2011) Santos <i>et al.</i> (2018)
Calcium (Ca) [mg/g DM]	Calcium Oxalates = anti-herbivore defense	Finley (1999) Hanley <i>et al.</i> (2007)
Iron (Fe) [mg/g DM]	Iron accumulation reduces leaf herbivory	Ribeiro <i>et al.</i> (2017)
Mangan (Mn) [mg/g DM]	Mangan accumulation reduces leaf herbivory	Ribeiro <i>et al.</i> (2017)

Carbon to Nitrogen Ratio (CN)	Associated to herbivory in numerous studies, therefore here tested despite weak correlation with specific leaf area	e.g., Njovu <i>et al.</i> (2019) Busch and Phelan (1999)
Nitrogen to Phosphorus ratio (NP)	High Nitrogen to Phosphorus ratio associated to low herbivory levels	Njovu <i>et al.</i> (2019)

Table A4: Spearman correlation coefficient between monthly community herbivory [%] and both mean monthly temperature [°C] and monthly precipitation [mm]. Non-significant correlation coefficients in grey.

Timeshift in months	temperature	precipitation
0	0.25	0.05
1	0.33	0
2	0.33	-0.11
3	0.37	-0.05
4	0.35	-0.19
5	0.34	-0.22
6	0.28	-0.39
7	0.15	-0.38
8	0.06	-0.32
9	0.06	0.01
10	0.07	0.01
11	0.15	0.12

Tab. A5: Mean community leaf herbivory [%] and leaf traits significantly influencing leaf herbivory for each elevational level [m a.s.l.]. SLA = Community weighted mean specific leaf area [cm²/g]; Ca = Community weighted mean leaf calcium concentration [mg/g]; Fe = Community weighted mean leaf iron concentration [mg/g]; MPD = Mean pairwise distance.

Elevation	Herbivory	SLA	Ca	Fe	MPD
1000	3.4 %	117	9.87	0.09	-1.13
2000	3.5 %	77	4.48	0.06	-0.03
3000	2.5 %	57	6.34	0.06	0.16

Tab. A6: Variables occurring in the minimal adequate models used for the structural equation model to analyze their impacts on community leaf herbivory [%]. SLA = Community weighted mean specific leaf area [cm²/g]; Ca = Community weighted mean leaf calcium concentration [mg/g DM]; Fe = Community weighted mean leaf iron concentration [mg/g DM]; MPD = Mean pairwise phylogenetic distance, temperature = monthly mean [°C]; precipitation = monthly mean [°C]; elevation = elevational level [m a.s.l.].

Response	Predictors	Random effect
MPD	Temperature, Precipitation	Elevation
SLA	MPD	Elevation
Ca	MPD, SLA	Elevation
Fe	MPD, SLA, Ca	Elevation
Herbivory	SLA, Ca, Fe	Month

Tab. A7: Monthly percentage of community herbivory [%] per and across elevational levels. Minimum and maximum herbivory values are in bold.

Month	1000 m a.s.l.	2000 m a.s.l.	3000 m a.s.l.	Mean across elevation
January	2.12	2.52	2.09	2.22
February	4.44	3.4	2.67	3.33
March	3.93	3.8	3.05	3.59
April	9.57	6.66	4.64	7.05
May	4.52	5.75	3.62	4.54
June	4.71	2.68	2.84	3.23
July	2.45	2.47	1.73	2.21
August	3.14	3.1	2.63	2.98
September	2.23	2.89	1.64	2.27
October	1.77	3.01	1.82	2.27
November	1.89	2.88	1.65	2.23
December	2.74	3.72	2.19	2.85

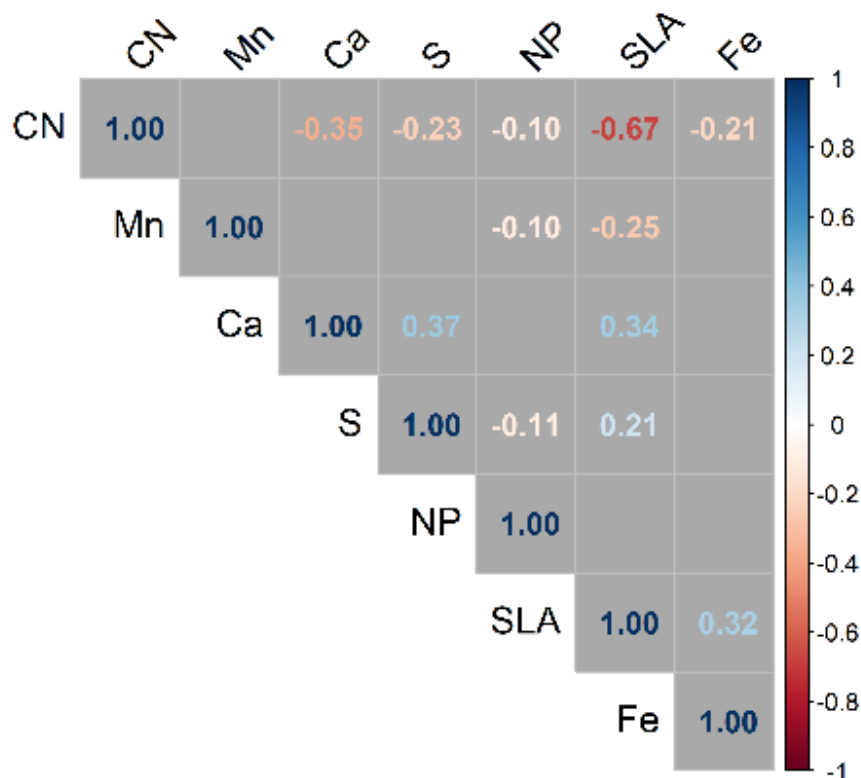


Figure A1: Pearson’s correlation coefficients matrix of measured leaf traits. Blank spaces indicate a non-significant correlation between two leaf traits. For abbreviation explanations of leaf traits see Tab. A1 column “Trait”.